Supporting Information for:

A Fibril-Like Assembly of Oligomers of a Peptide Derived from β-Amyloid.

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This PDF file includes:

Materials and Methods:

Peptides **3–6** were prepared and studied as the trifluoroacetate (TFA) salts. ^{1}H NMR 1D, TOCSY, NOESY, ROESY, and DOSY experiments for peptides **3–6** were carried out as previously described. [1]

*Synthesis of macrocyclic β-sheet peptides **3–6**.*

Macrocyclic β-sheet peptides **3–6** were synthesized using procedures previously reported for the synthesis of **1** and of other macrocyclic β-sheet peptides.[2,3,4] Boc-Orn(Fmoc)-OH was used to introduce the δ-linked ornithine turn units. Fmoc-Hao-OH was used to introduce the unnatural amino acid Hao.[3,5] Standard Fmoc-protected amino acids were used to introduce the other residues: Fmoc-Ala-OH, Fmoc-Asp(OtBu)-OH, Fmoc-Gln(Trt)-OH, Fmoc-Glu(OtBu)-OH, Fmoc-Leu-OH, Fmoc-Lys(Boc)-OH, Fmoc-Phe-OH, Fmoc-Phe(*p*-iodo)-OH, Fmoc-Ser(OtBu)-OH, Fmoc-Thr(OtBu)-OH, Fmoc-Tyr(OtBu)-OH, and Fmoc-Val-OH.

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1 Pham, J. D.; Demeler, B.; Nowick, J. S. *J. Am. Chem. Soc.* **2014**, *136*, 5432–5442.

2 Pham, J. D.; Chim, N.; Goulding, C. W.; Nowick, J. S. *J. Am. Chem. Soc.* **2013**, *135*, 12460–12467.

3 Cheng, P.-N.; Nowick, J. S. *J. Org. Chem.* **2011**, *76*, 3166–3173.

4 Cheng, P.-N.; Liu, C.; Zhao, M.; Eisenberg, D.; Nowick, J. S. *Nat. Chem.* **2012**, *4*, 927–933.

5 Nowick, J. S.; Chung, D. M.; Maitra, K.; Maitra, S.; Stigers, K. D.; Sun, Y. J. *J. Am. Chem. Soc.* **2000**, *122*, 7654–7661.

X-ray diffraction data collection, processing, and structure refinement.

Crystals of macrocyclic β-sheet peptide **3** were flash-frozen in liquid nitrogen prior to data collection. Diffraction data for macrocyclic β-sheet peptide **3** were collected on beamline 7-1 at the Stanford Synchrotron Radiation Lightsource (Stanford, CA) at 1.00 Å wavelength to a resolution 1.75 Å at 100 K. [Data were collected at 1.00 Å wavelength to take advantage of the maximum flux of the beamline and obtain a reasonable anomalous signal from the iodine groups (*f*" = 3.3).] Data were collected over 180 degrees with a 0.5 degree oscillation. The data were integrated and scaled with XDS,[6] and merged with Aimless.[7] The space group was initially determined to be $C222_1$; after an analysis of the data with Xtriage in the PHENIX software suite,[8] it became apparent (multivariate *Z* score *L*-test > 3.5) that the data better fit a lower space group with pseudomerohedral twinning.[9] Data were reprocessed in the *C*2 space group and refined with the appropriate twin law (*-h*, *-k*, *l*).

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6 Kabsch, W. *Acta Crystallogr. Sect. D: Biol. Crystallogr.* **2010**, *66*, 125–132.

7 (a) Evans, P. *Acta Crystallogr. Sect. D: Biol. Crystallogr.* **2006**, *62*, 72–82. (b) Evans, P. R.; Murshudov, G. N. *Acta Crystallogr. Sect. D: Biol. Crystallogr.* **2013**, *69*, 1204–1214.

8 Adams, P. D.; Afonine, P. V.; Bunkoczi, G.; Chen, V. B.; Davis, I. W.; Echols, N.; Headd, J. J.; Hung, L. W.; Kapral, G. J.; Grosse-Kunstleve, R. W.; McCoy, A. J.; Moriarty, N. W.; Oeffner, R.; Read, R. J.; Richardson, D. C.; Richardson, J. S.; Terwilliger, T. C.; Zwart, P. H. *Acta Crystallogr. Sect. D: Biol. Crystallogr.* **2010**, *66*, 213–221.

9 Larsen, N. A.; Heine, A.; de Prada, P.; Redwan, E.-R.; Yeates, T. O.; Landry, D. W.; Wilson, I. A. *Acta Crystallogr. Sect. D: Biol. Crystallogr.* **2002**, *58*, 2055–2059.

Initial positions for the iodine groups were determined using hybrid substructure search (HySS) in the PHENIX software suite.[8,10] Coordinates for the iodine groups determined by HySS were used with Autosol to generate the initial electron density map. Two macrocyclic β-sheet peptides **3** and two 2-methyl-2,4-pentanediol molecules were found in the asymmetric unit. Iterative rounds of refinement and model building were done with phenix.refine and Coot,[11] respectively. Each structure during the iterative model building was refined with riding hydrogens, TLS parameters, anisotropic *B*-factors for 4-iodophenylalanine residues, and with the twin law (-*h*, -*k*, *l*). Statistics for the final refinement of macrocyclic β-sheet peptides **3** were R_{work} = 17.94% and R_{free} = 21.97%. The crystal structure was deposited to the Protein Data Bank (PDB) with PDB code 4Q8D.

PyMOL was used to generate images from the crystallographic data. A β-strand of three glycine residues (G3) was used to replace Hao in generating a cartoon of the $A\beta_{15–23}$ hybrid strand, QKLV-Hao-ED. Specifically, the pdb coordinates for the unnatural amino acid Hao were used to generate triglycine segments.

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[10] Grosse-Kunstleve, R. W.; Adams, P. D. *Acta Crystallogr. Sect. D: Biol. Crystallogr.* **2003**, *59*, 1966–1973.

[11] Emsley, P.; Lohkamp, B.; Scott, W. G.; Cowtan, K. *Acta Crystallogr. Sect. D: Biol. Crystallogr.* **2010**, *66*, 486–501.

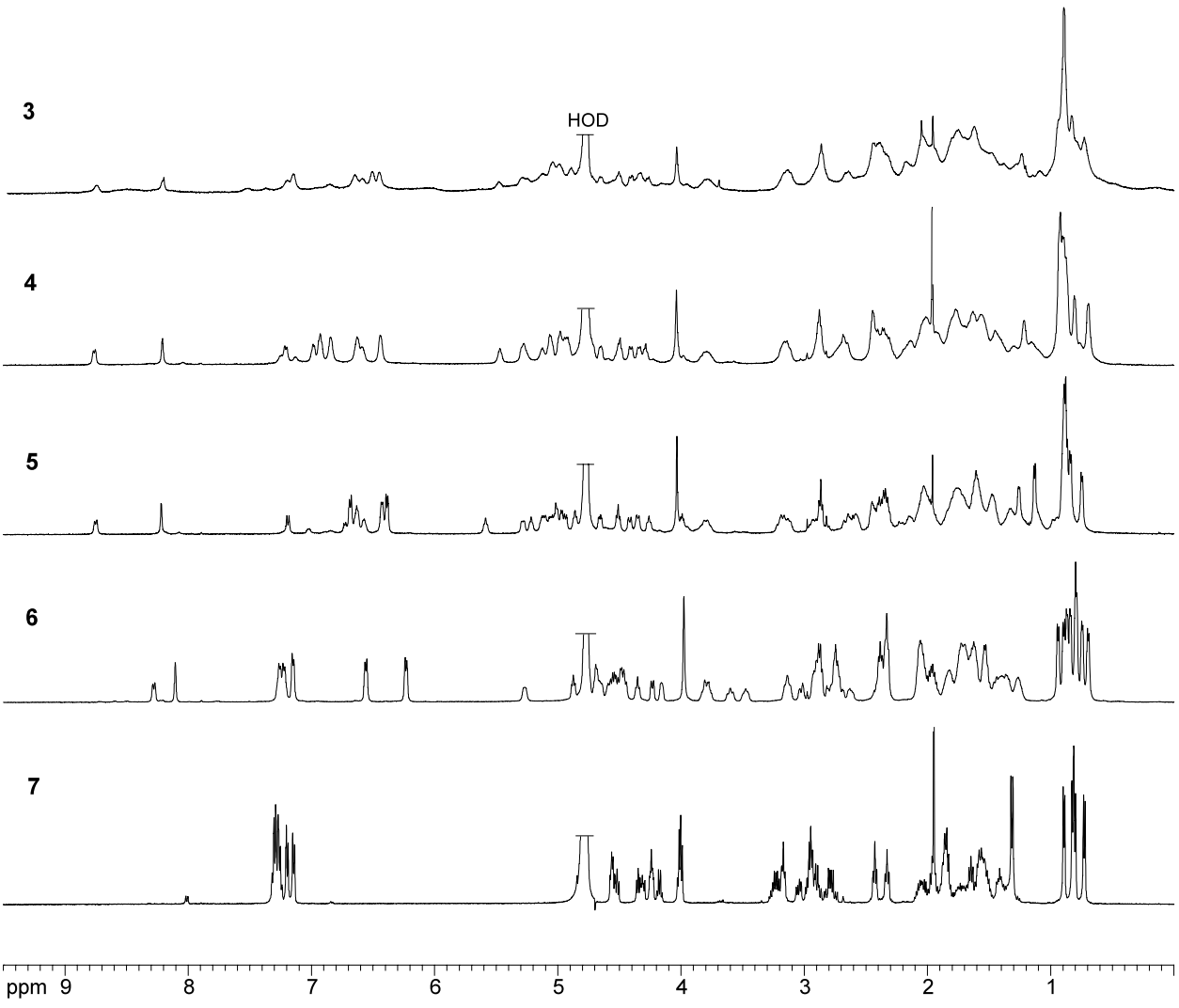
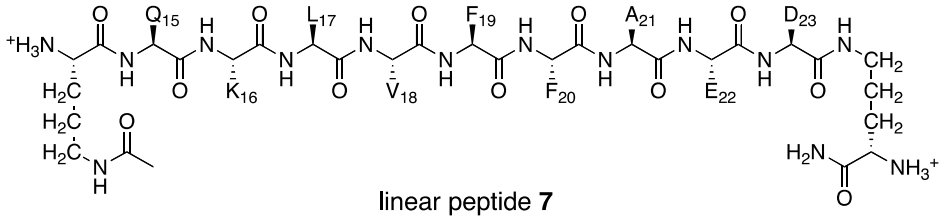


Figure S1. ^{1}H NMR spectra of macrocyclic β-sheet peptides at 2.0 mM at 298 K in D_2O at 500 MHz: **3** (tetramer predominates), **4** (tetramer predominates), **5** (tetramer predominates), and **6** (monomer predominates). The ^{1}H NMR spectrum of linear peptide **7** (1.2 mM at 298 K in D_2O at 500 MHz) is provided for comparison, to show the spectrum of an unstructured QKLVFFAED peptide.[1]



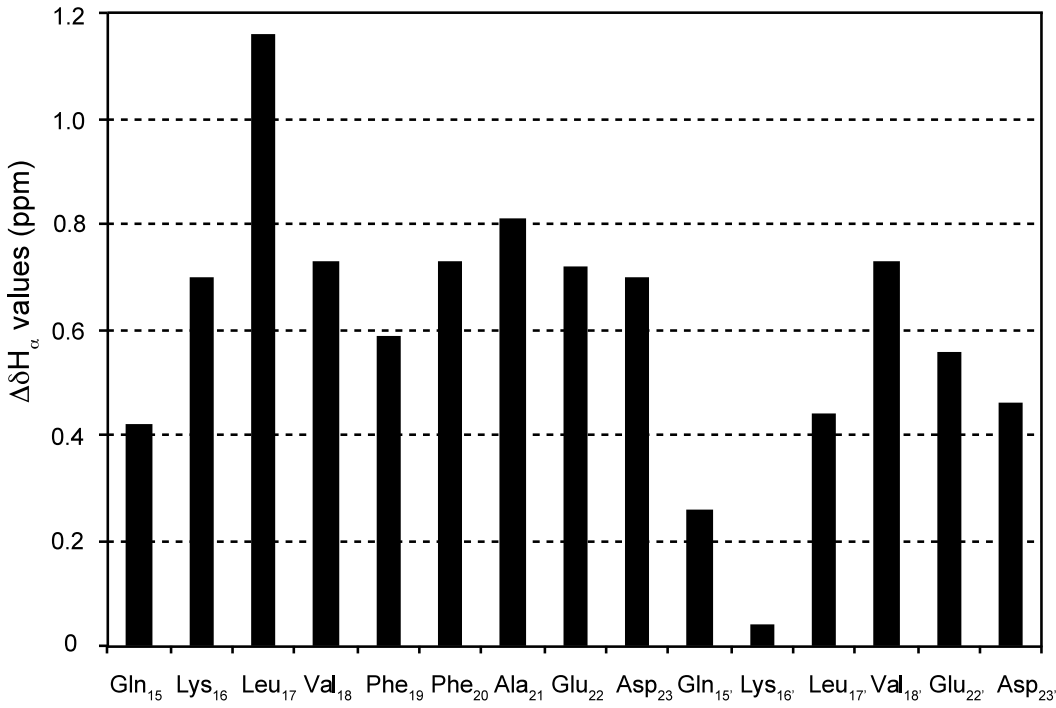


Figure S2. Downfield shifting of the ^{1}H NMR α-proton resonances of the tetramer of macrocyclic β-sheet peptide **4** relative to linear peptide **7**[1]. The ^{1}H NMR spectrum of **4** was recorded at 2.0 mM in D_2O at 500 MHz and 298 K. Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, and Gln_{15} vs. $Gln_{15'}$ are arbitrary.

Table S1. Magnetic Anisotropies of the δ-Protons of the δ-Linked Ornithine Turn Units of Peptides 3–6 at 2.0 mM in D_2O at 298 K and 500 MHz

peptide	$^{\delta}Orn_1$[c] Δδ (ppm)	$^{\delta}Orn_2$[c] Δδ (ppm)	folding
3[a]	0.63	0.69	folded tetramer
4[a]	0.60	0.69	folded tetramer
5[a]	0.62	0.70	folded tetramer
6[b]	0.46	0.34	partially folded monomer

[a]Oligomer at 2.0 mM. [b] Monomer at 2.0 mM. [c]Assignment of $^{\delta}Orn_1$ vs. $^{\delta}Orn_2$ is arbitrary.

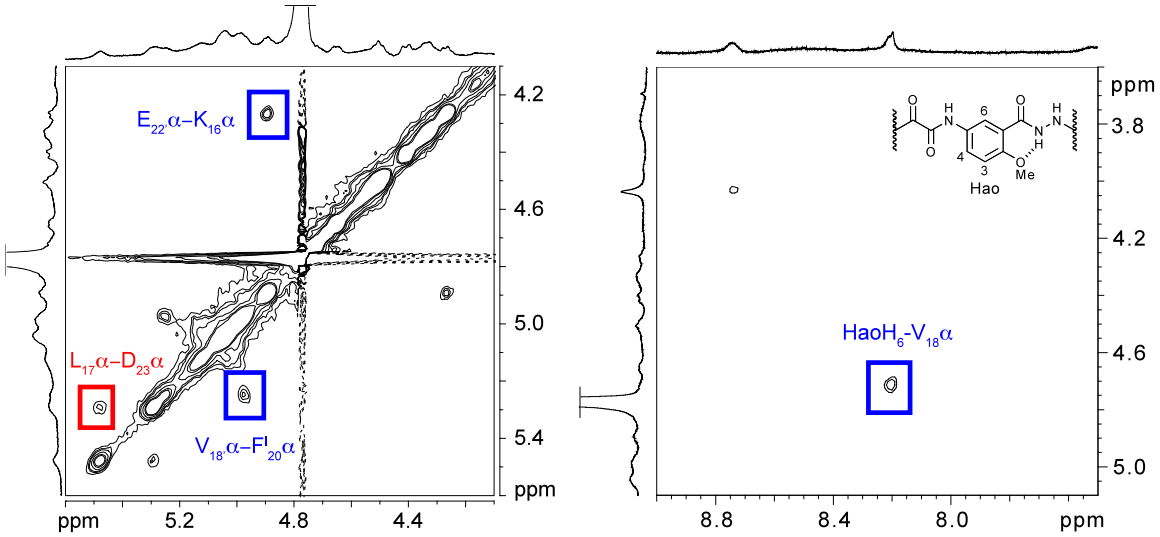


Figure S3. Selected expansions of the NOESY spectrum of macrocyclic β-sheet peptide **3** at 2.0 mM in D_2O at 500 MHz and 298 K. Key intermolecular interstrand NOEs associated with dimerization are highlighted in red; key intramolecular interstrand NOEs associated with folding are highlighted in blue.

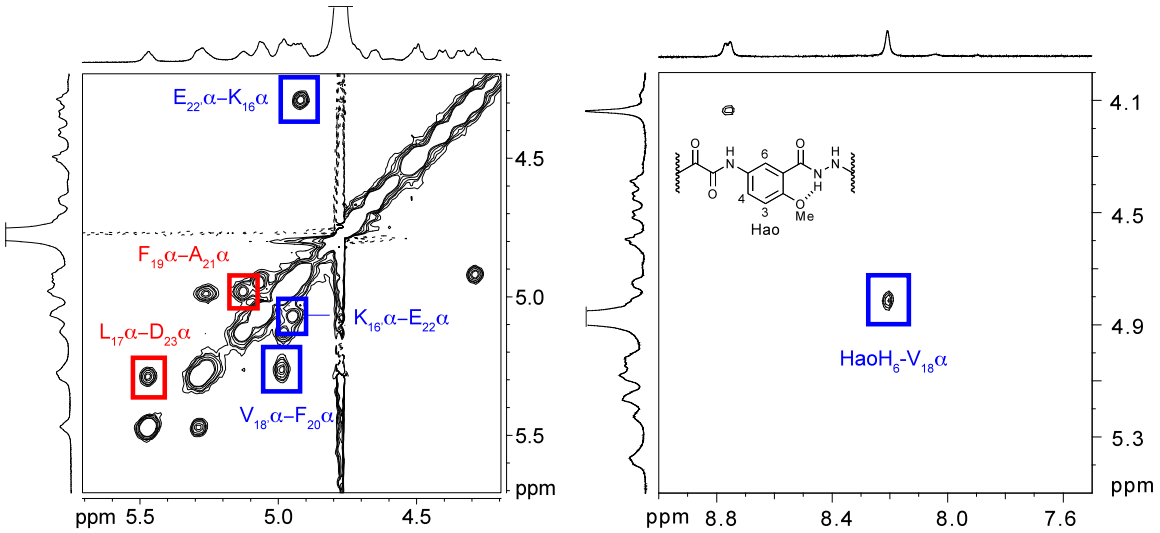


Figure S4. Selected expansions of the NOESY spectrum of macrocyclic β-sheet peptide **4** at 2.0 mM in D_2O at 500 MHz and 298 K. Key intermolecular interstrand NOEs associated with dimerization are highlighted in red; key intramolecular interstrand NOEs associated with folding are highlighted in blue.

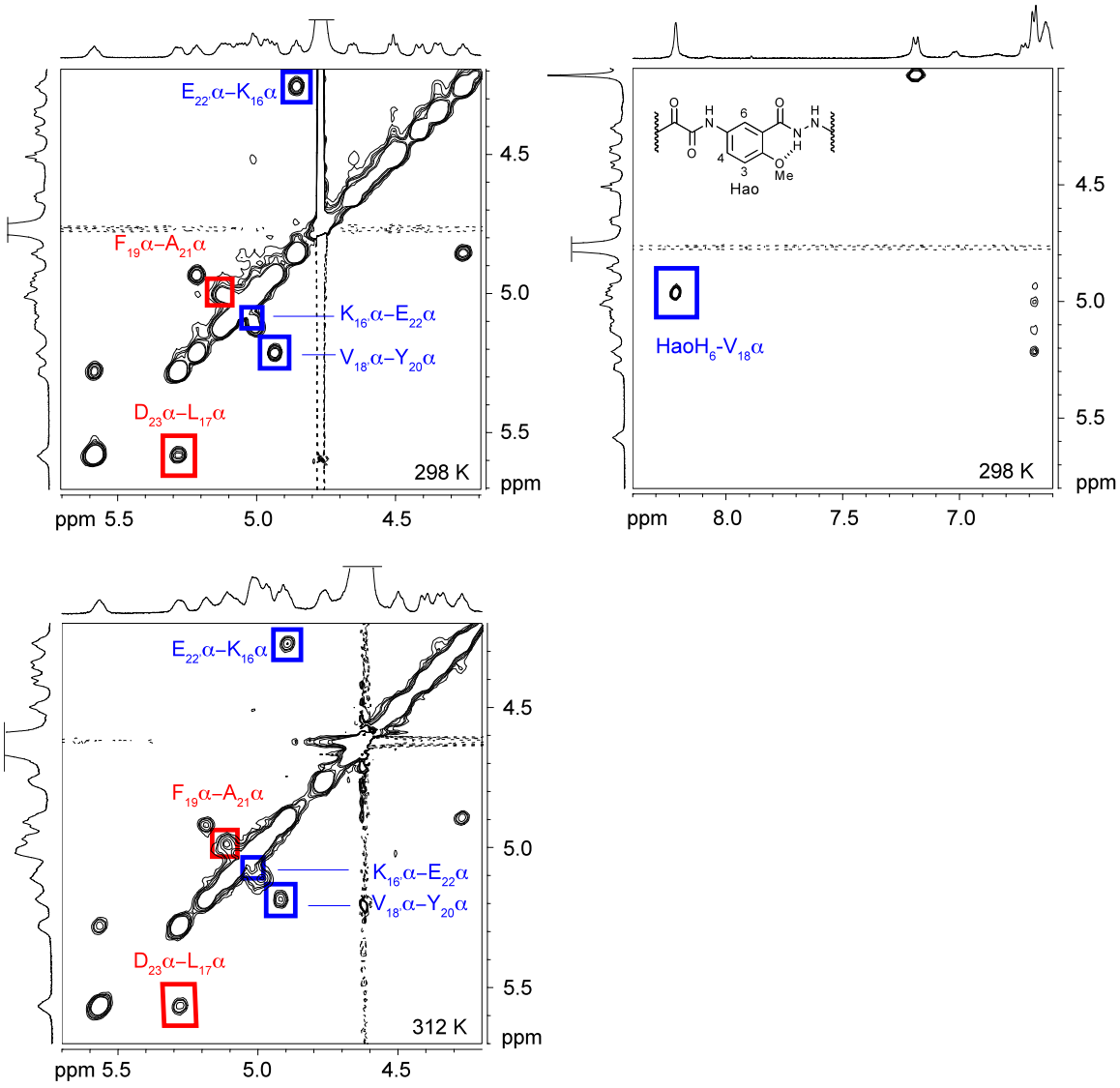
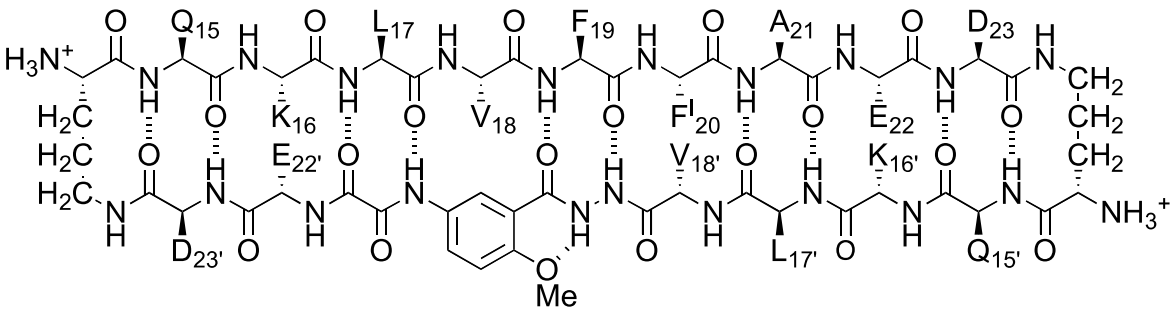


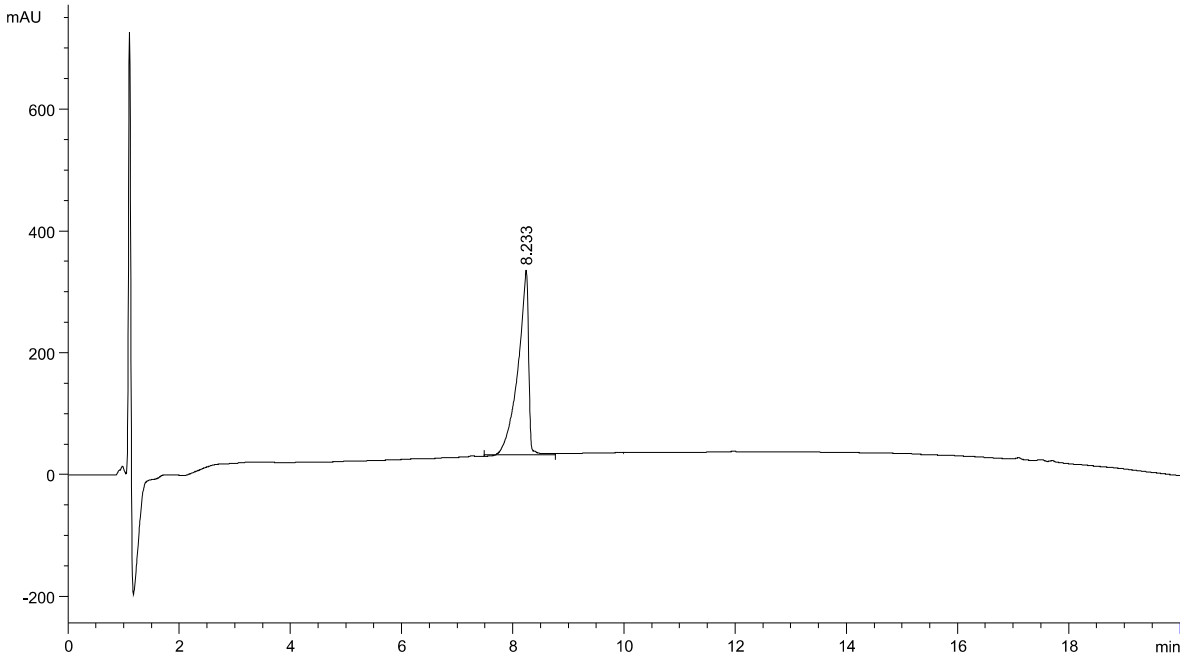
Figure S5. Selected expansions of the NOESY spectrum of macrocyclic β-sheet peptide **5** at 2.0 mM in D_2O at 500 MHz and at 298 K and 312 K. Key intermolecular interstrand NOEs associated with dimerization are highlighted in red; key intramolecular interstrand NOEs associated with folding are highlighted in blue.



macrocyclic β-sheet peptide **3** (as the TFA salt)

molecular weight calculated for $C_{103}H_{155}IN_{26}O_{31} \cdot 4CF_3CO_2H$ (TFA salt of **3**): 2836.49
molecular weight calculated for $C_{103}H_{155}IN_{26}O_{31}$ (free base of **3**): 2380.39
exact mass calculated for $C_{103}H_{155}IN_{26}O_{31}$ (free base of **3**): 2379.04

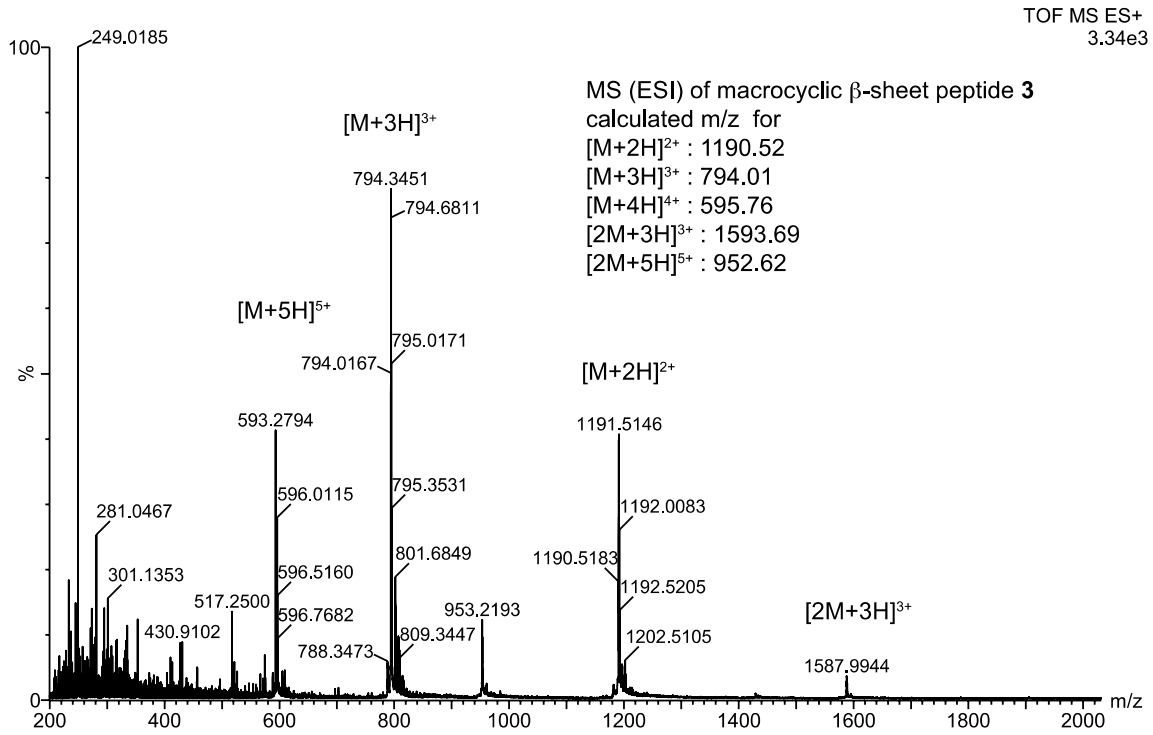
Analytical RP-HPLC of macrocyclic β-sheet **3**

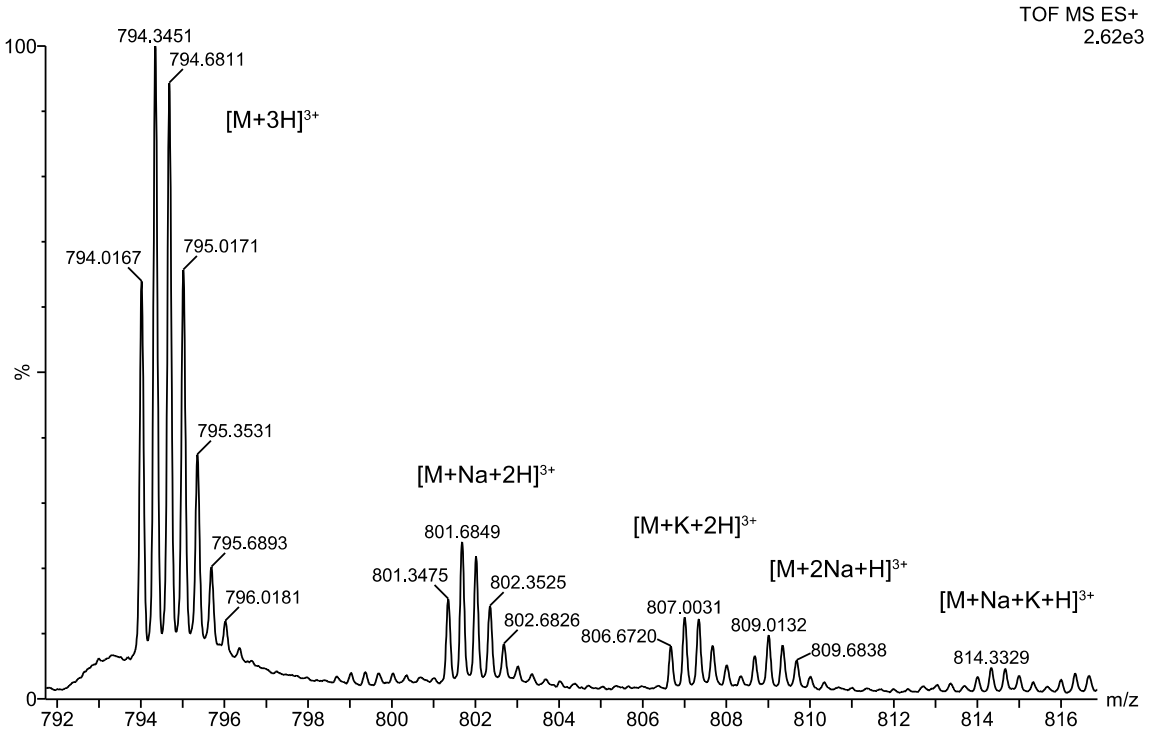


Signal 1: VWD1 A, Wavelength=214 nm

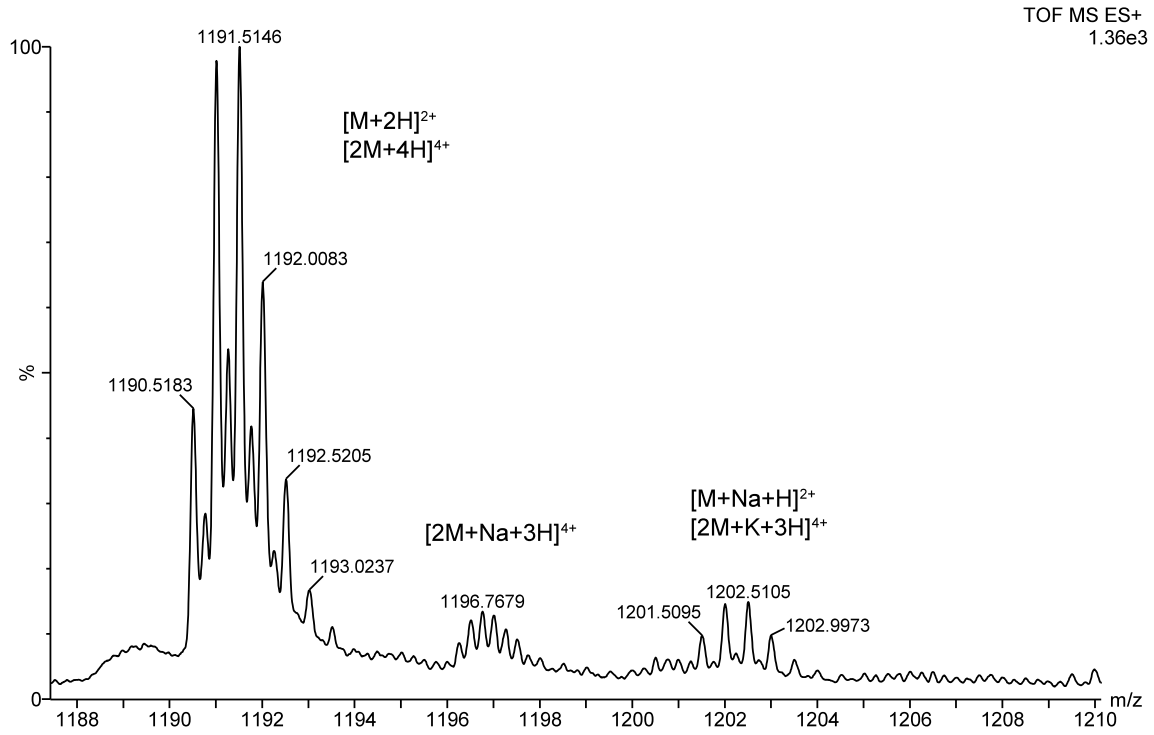
Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	8.233	MM	0.2195	3995.70361	303.33344	100.0000
Totals :				3995.70361	303.33344	

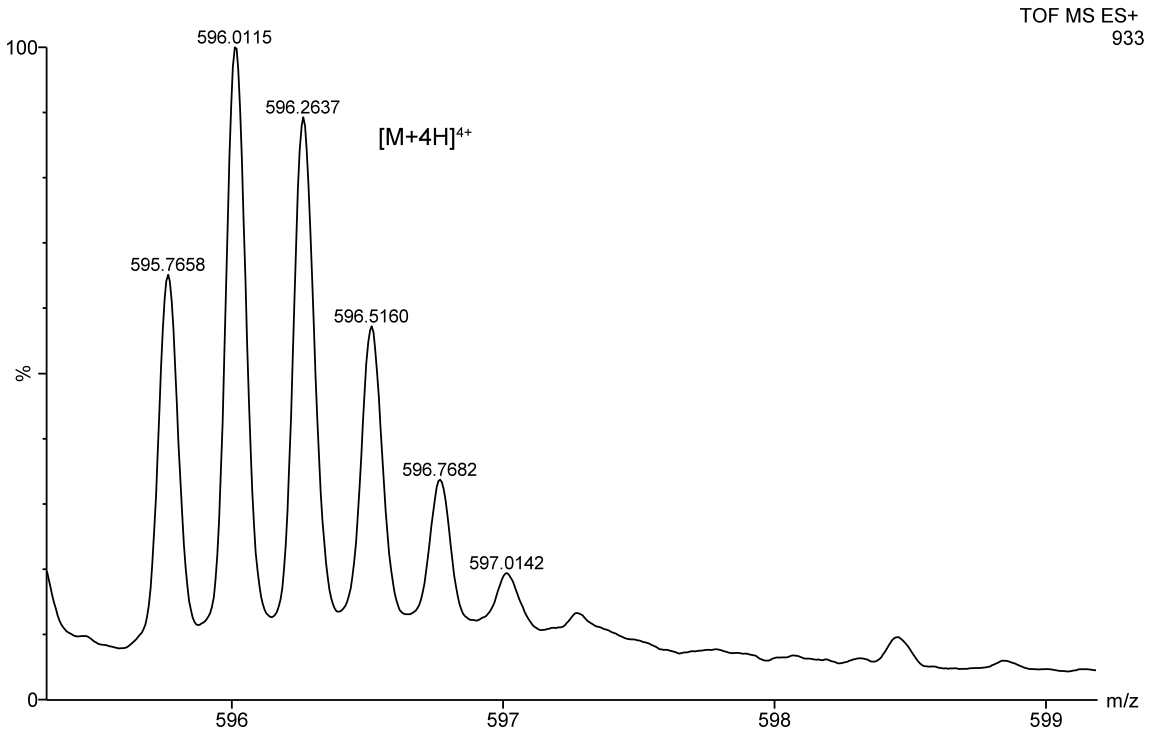
Macrocyclic β-sheet peptide **3**



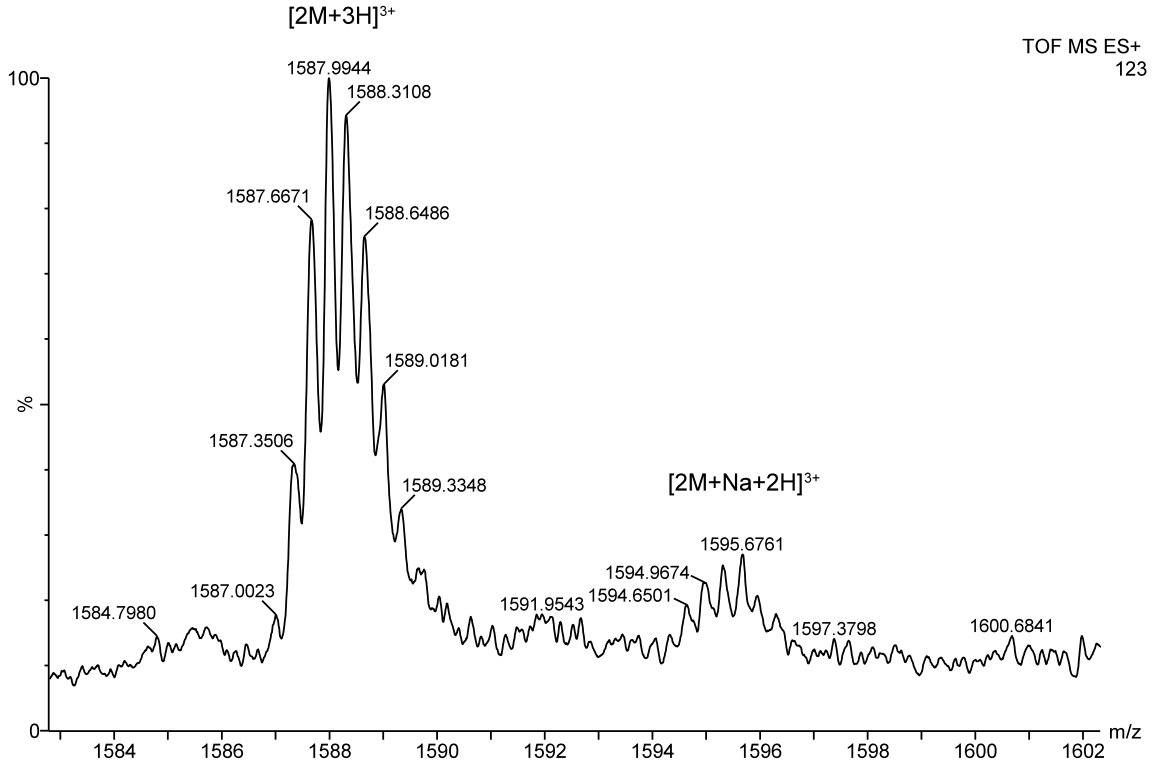


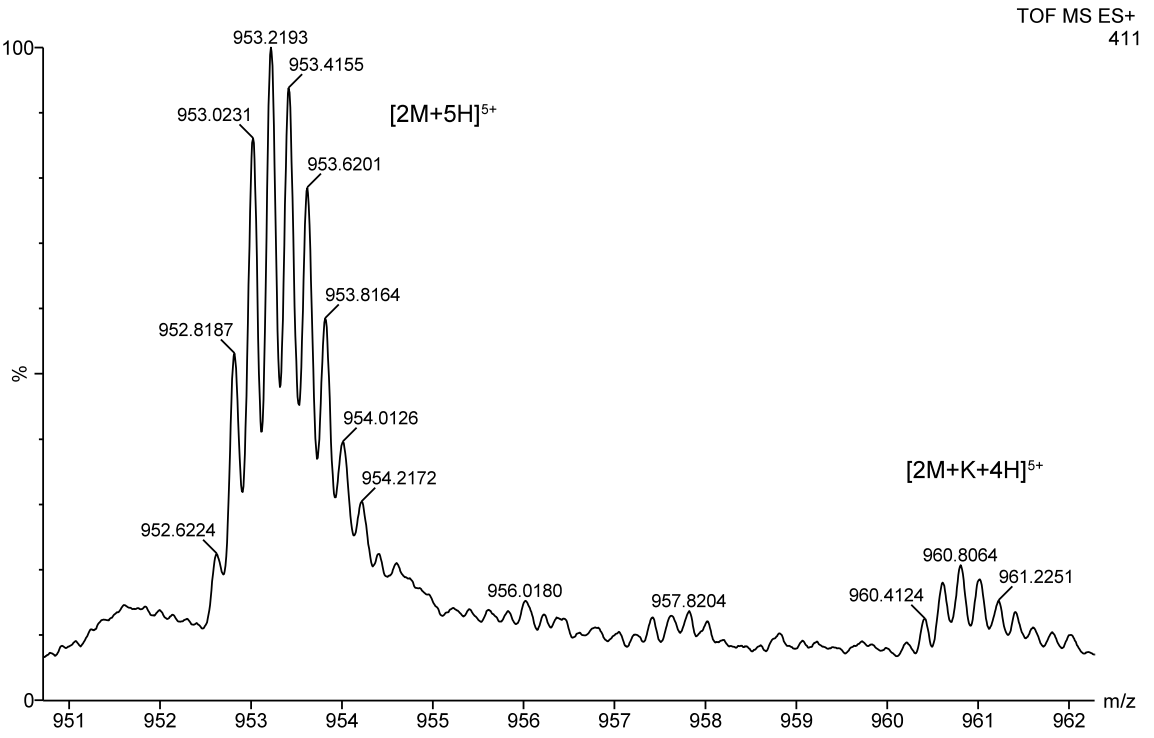
Macrocyclic β-sheet peptide **3**



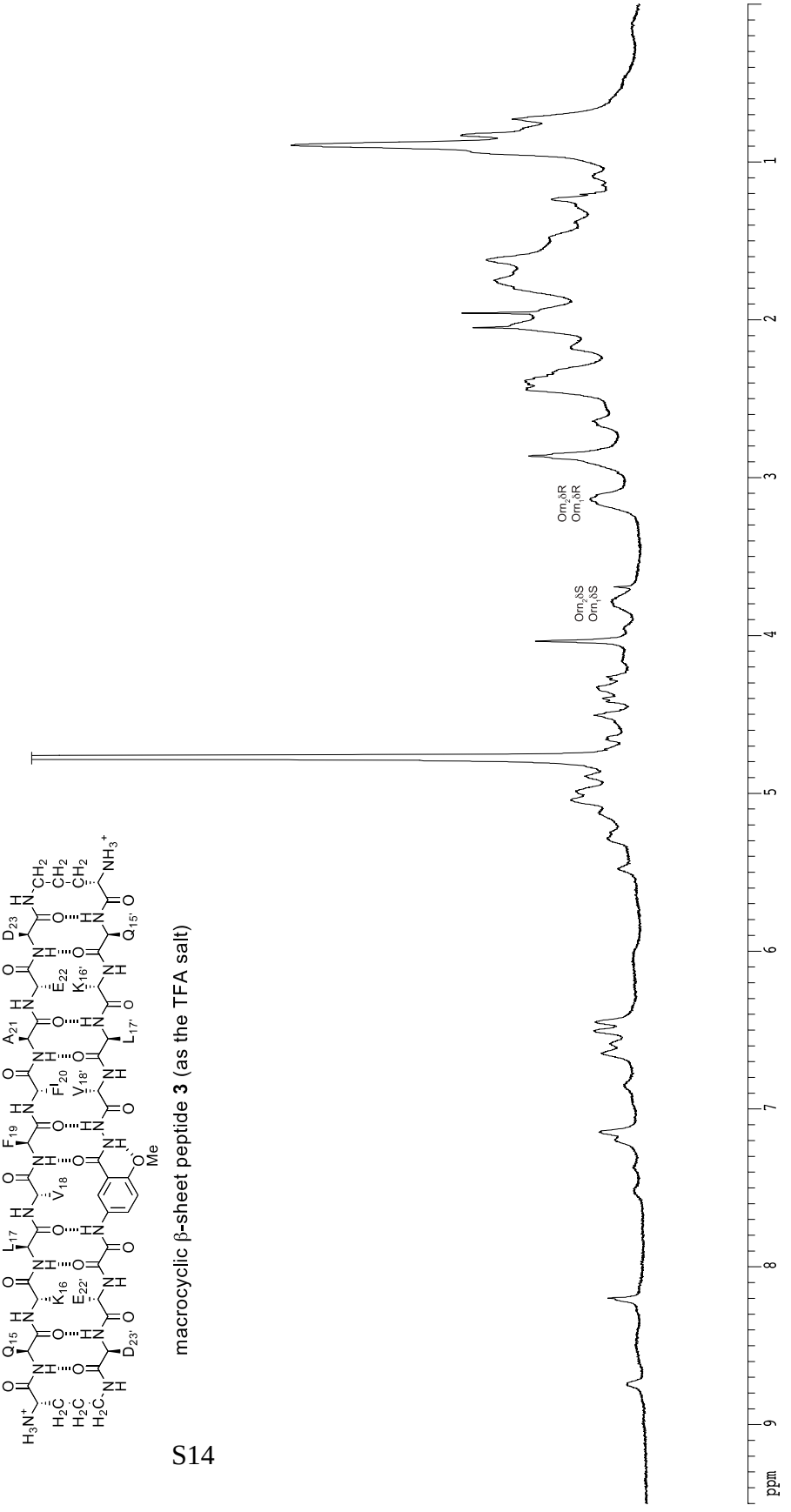


Macrocyclic β-sheet peptide **3**



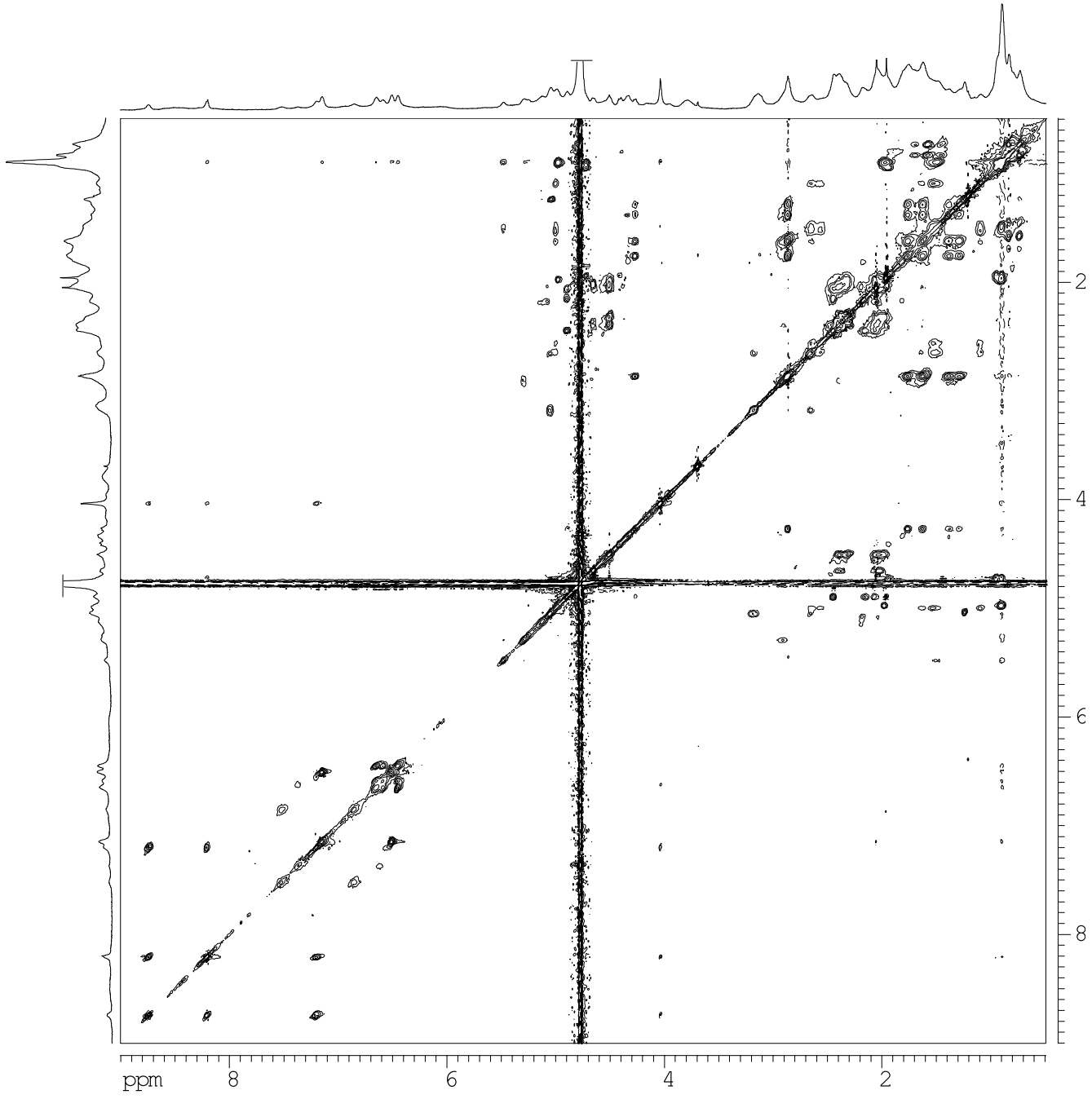


1D ^{1}H NMR spectrum of macrocyclic β-sheet **3**
2 mM in D_2O, 500 MHz, 298 K
tetramer predominates

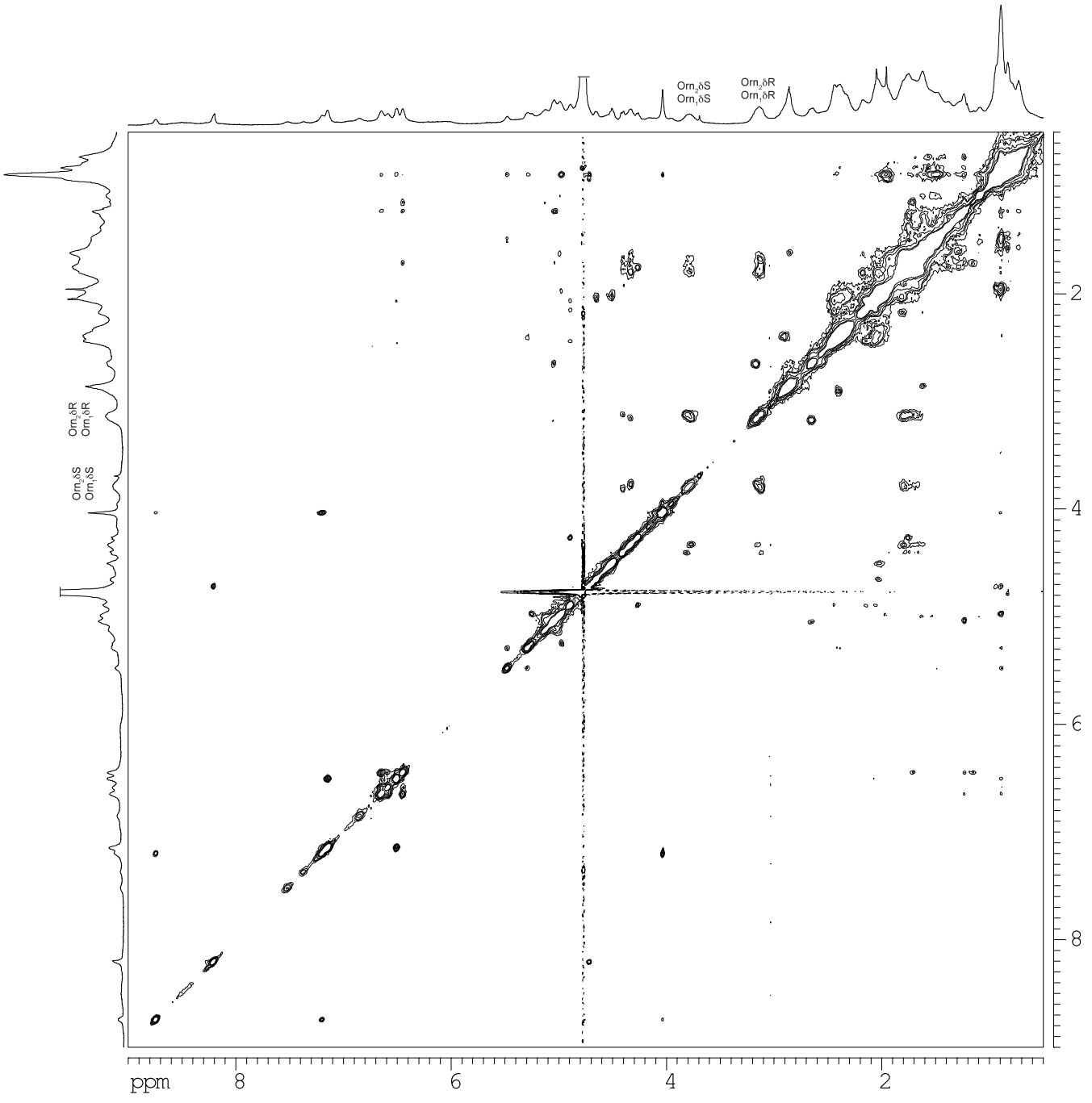


macrocyclic β-sheet peptide **3** (as the TFA salt)

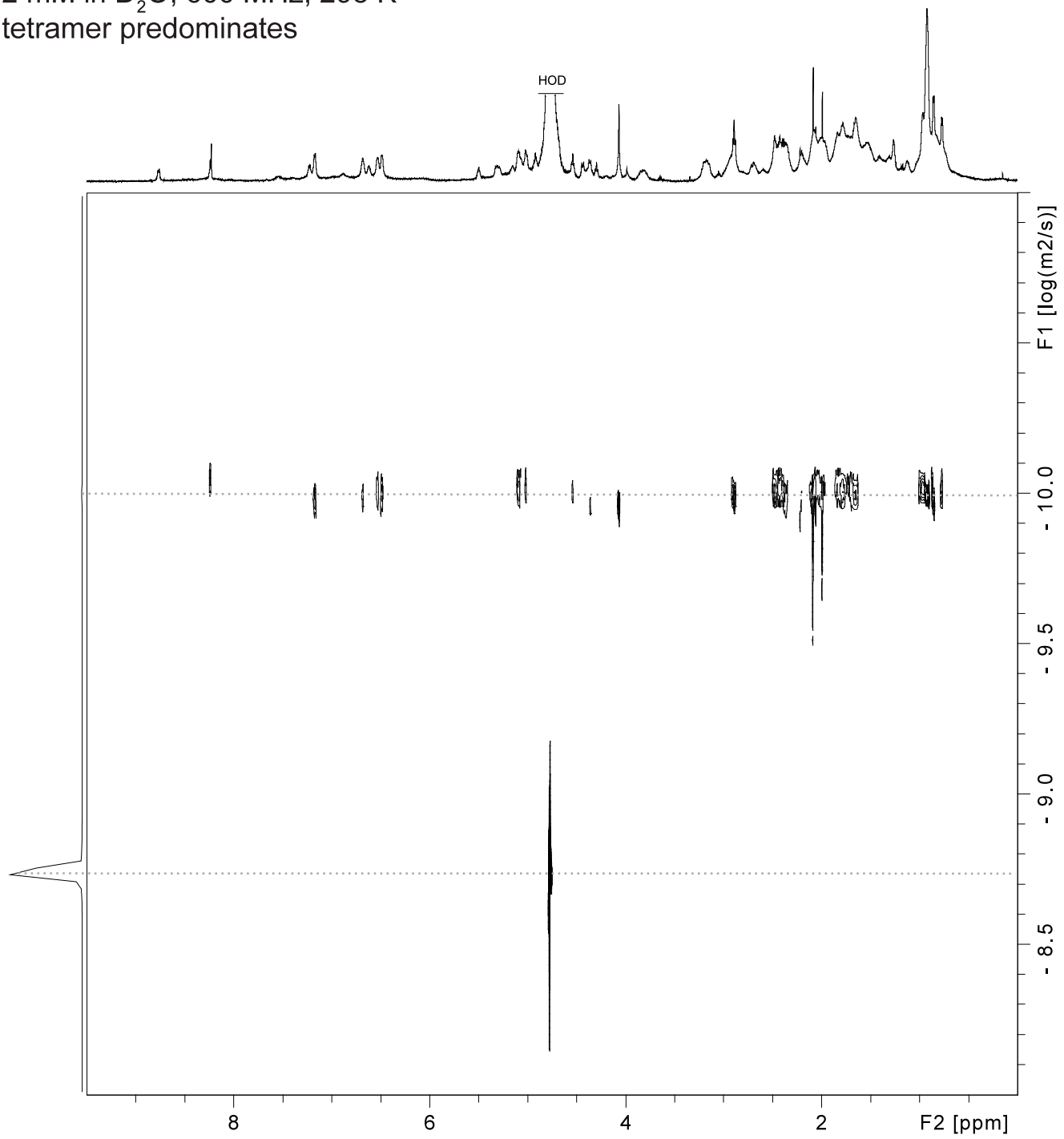
2D TOCSY spectrum of macrocyclic β-sheet **3**
2 mM in D_2O, 500 MHz, 298 K
150-ms spin-locking mixing time
tetramer predominates



2D NOESY spectrum of macrocyclic β-sheet **3**
2 mM in D_2O, 500 MHz, 298 K
200-ms spin-locking mixing time
tetramer predominates



2D DOSY spectrum of macrocyclic β-sheet **3**
2 mM in D_2O, 600 MHz, 298 K
tetramer predominates

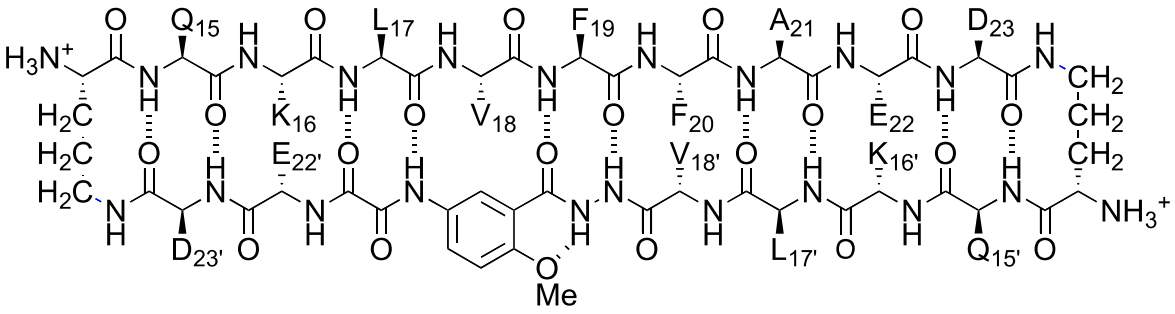


Calculation for peptide **3** at 2.0 mM

DC_{HOD} = 19.0 x 10^{-10} m^2/s [a]
log DC_{HOD} = -8.721

For peptide **3** tetramer, log DC (m^2/s) = -9.99(4), DC = $10^{-9.994}$ m^2/s = 10.1 x 10^{-11} m^2/s = 10.1 x 10^{-7} cm^2/s

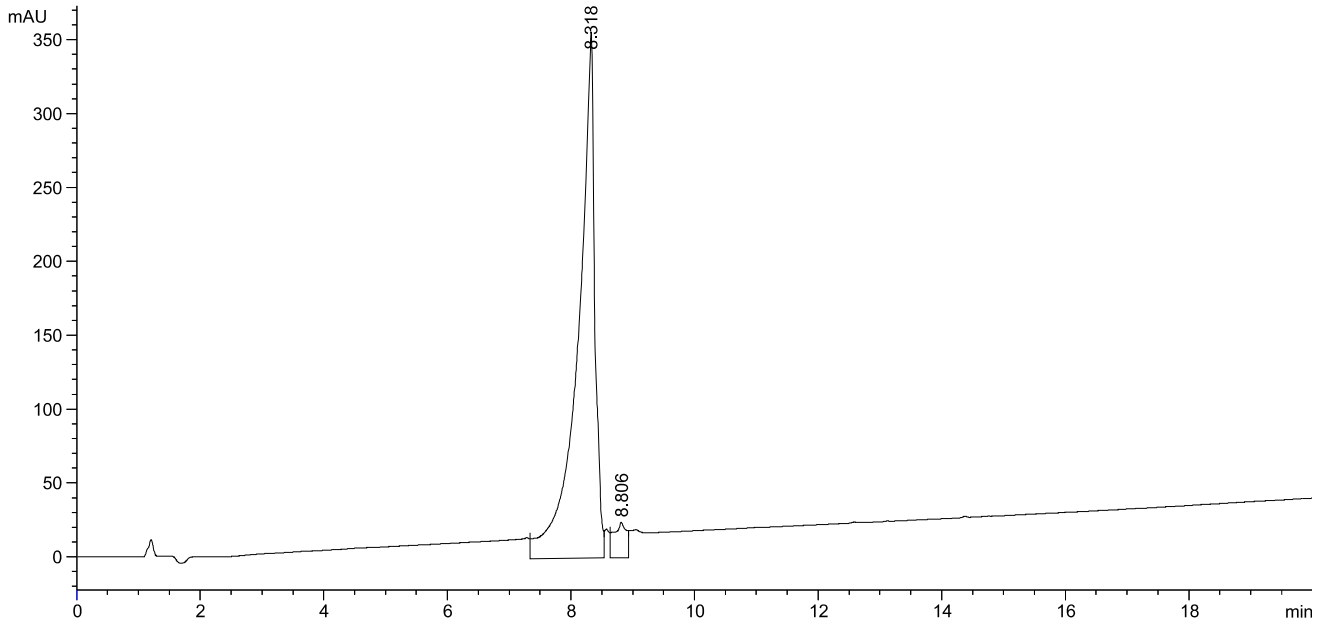
[a] Longsworth, L. G. J. Phys. Chem. 1960, 64, 1914–1917.



macrocyclic β-sheet peptide **4** (as the TFA salt)

molecular weight calculated for $C_{103}H_{156}N_{26}O_{31} \cdot 4CF_3CO_2H$ (TFA salt of **4**): 2710.59
molecular weight calculated for $C_{103}H_{156}N_{26}O_{31}$ (free base of **4**): 2254.50
exact mass calculated for $C_{103}H_{156}N_{26}O_{31}$ (free base of **4**): 2253.14

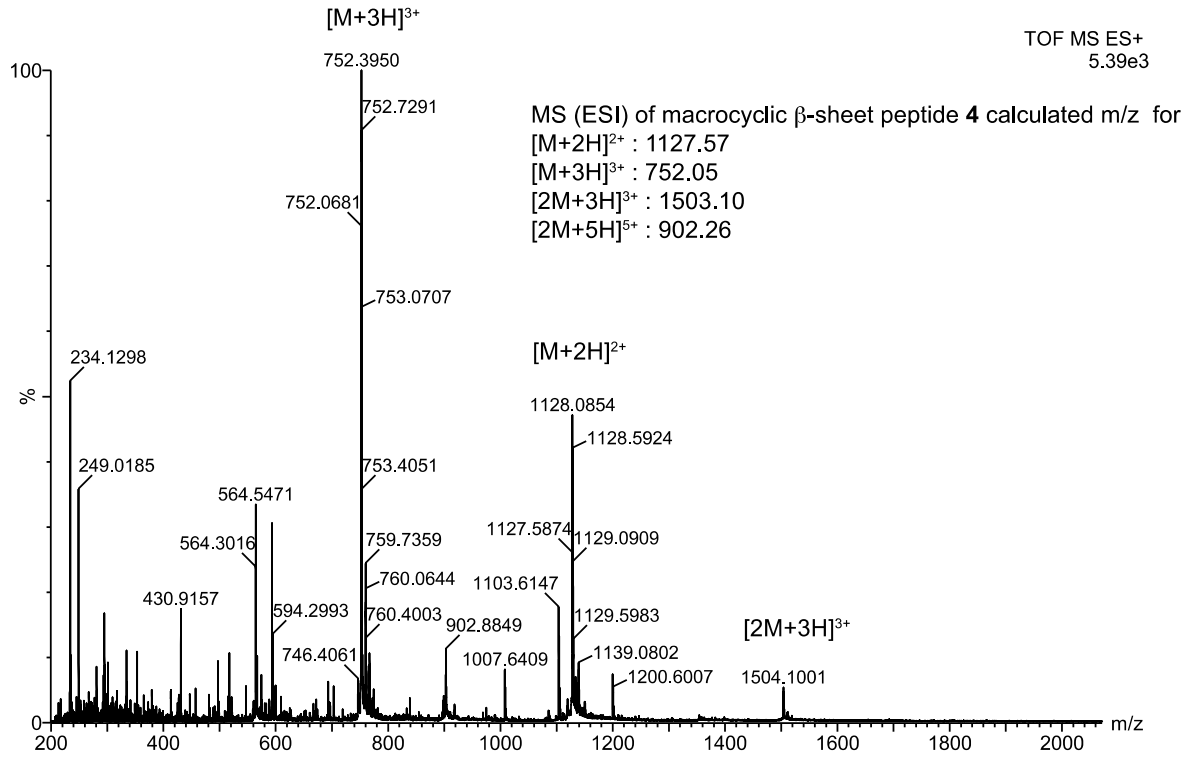
Analytical RP-HPLC of macrocyclic β-peptide **4**

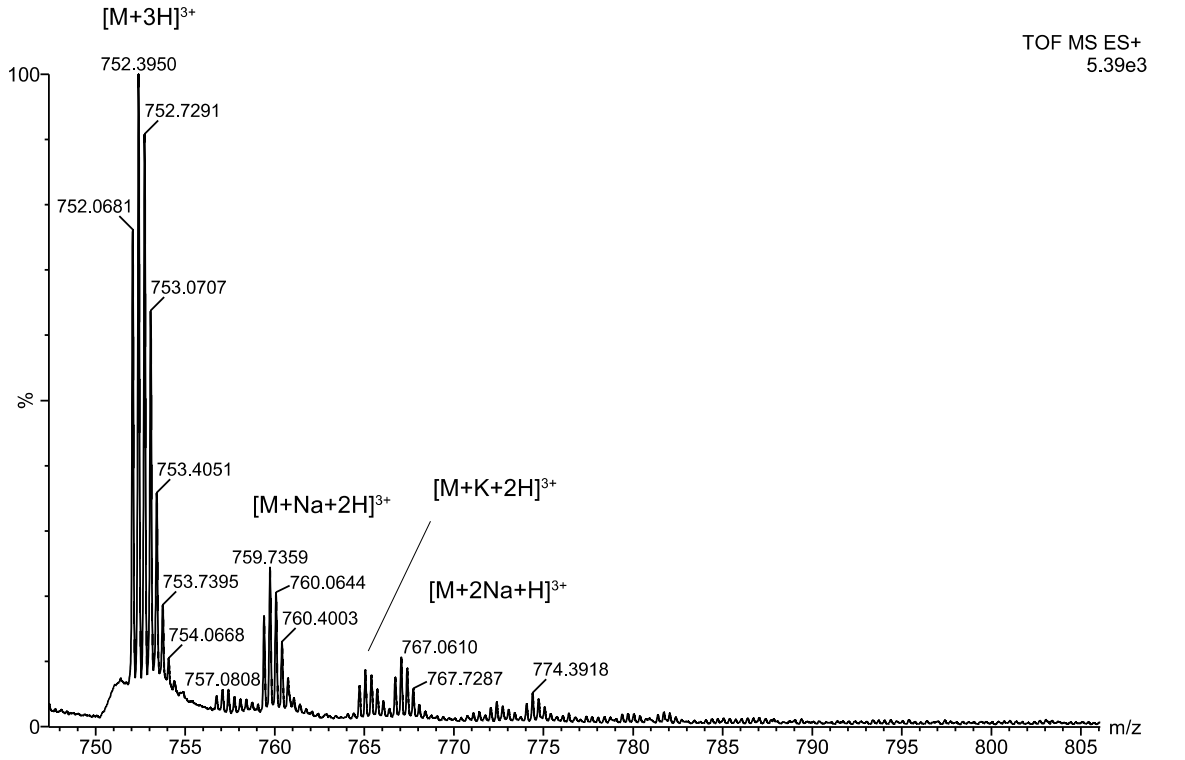


Signal 1: VWD1 A, Wavelength=254 nm

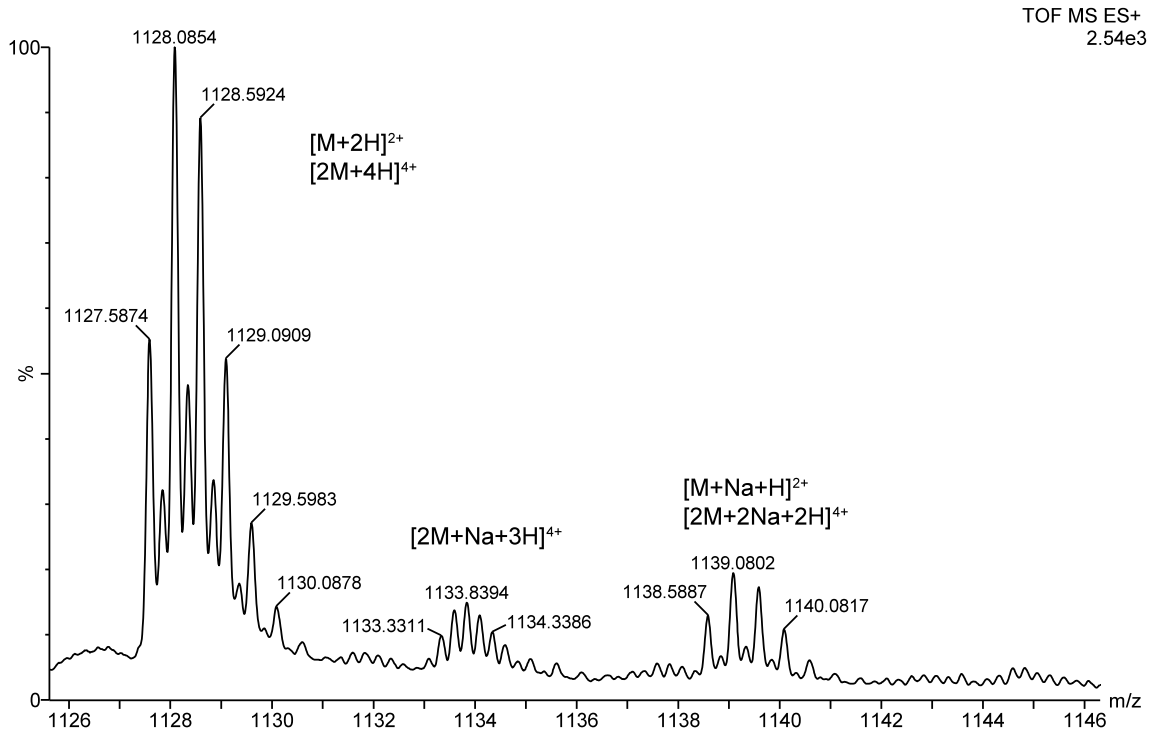
Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	8.318	VV	0.2395	6692.11328	356.28714	95.1129
2	8.806	VV	0.1819	343.85294	23.96618	4.8871

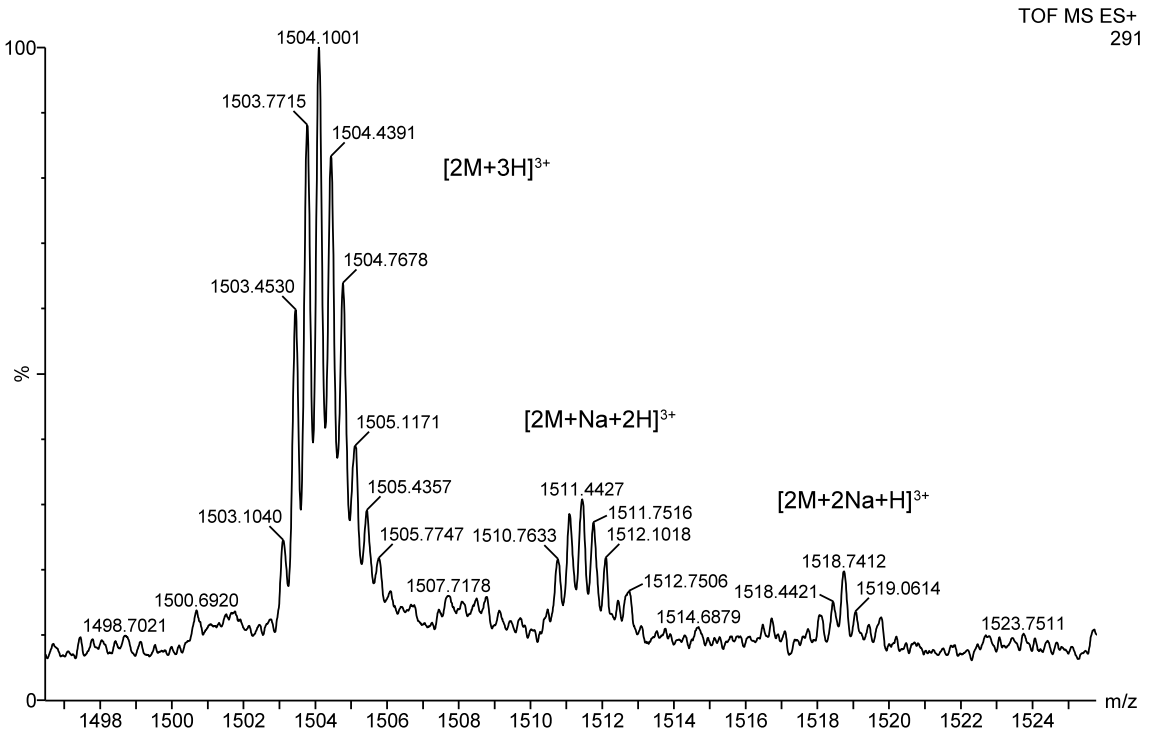
Macrocyclic β-sheet peptide **4**



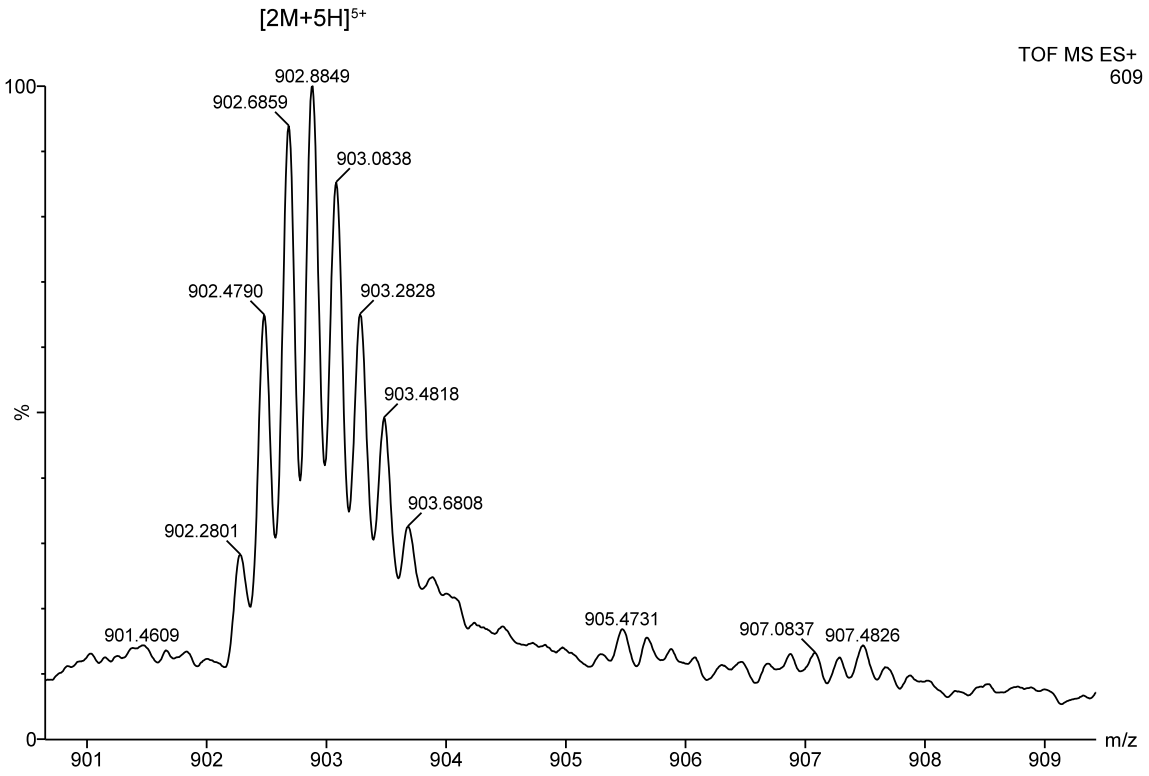


Macrocyclic β-sheet peptide **4**



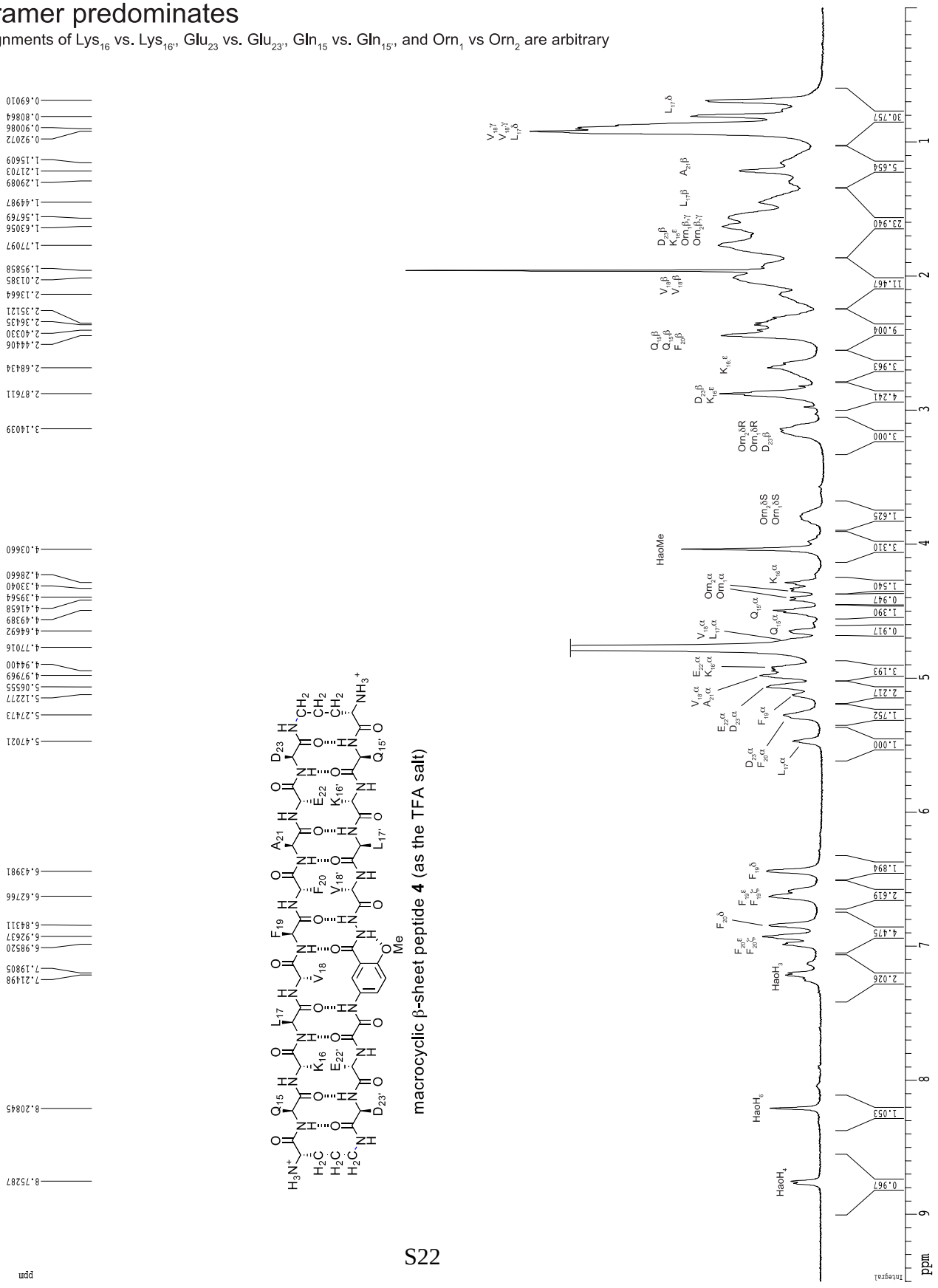


Macrocyclic β-sheet peptide **4**



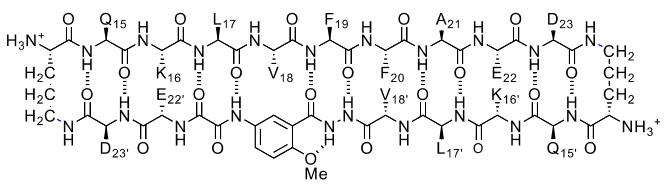
1D ^{1}H NMR spectrum of macrocyclic β-sheet **4**
2 mM in D_2O, 500 MHz, 298 K
tetramer predominates

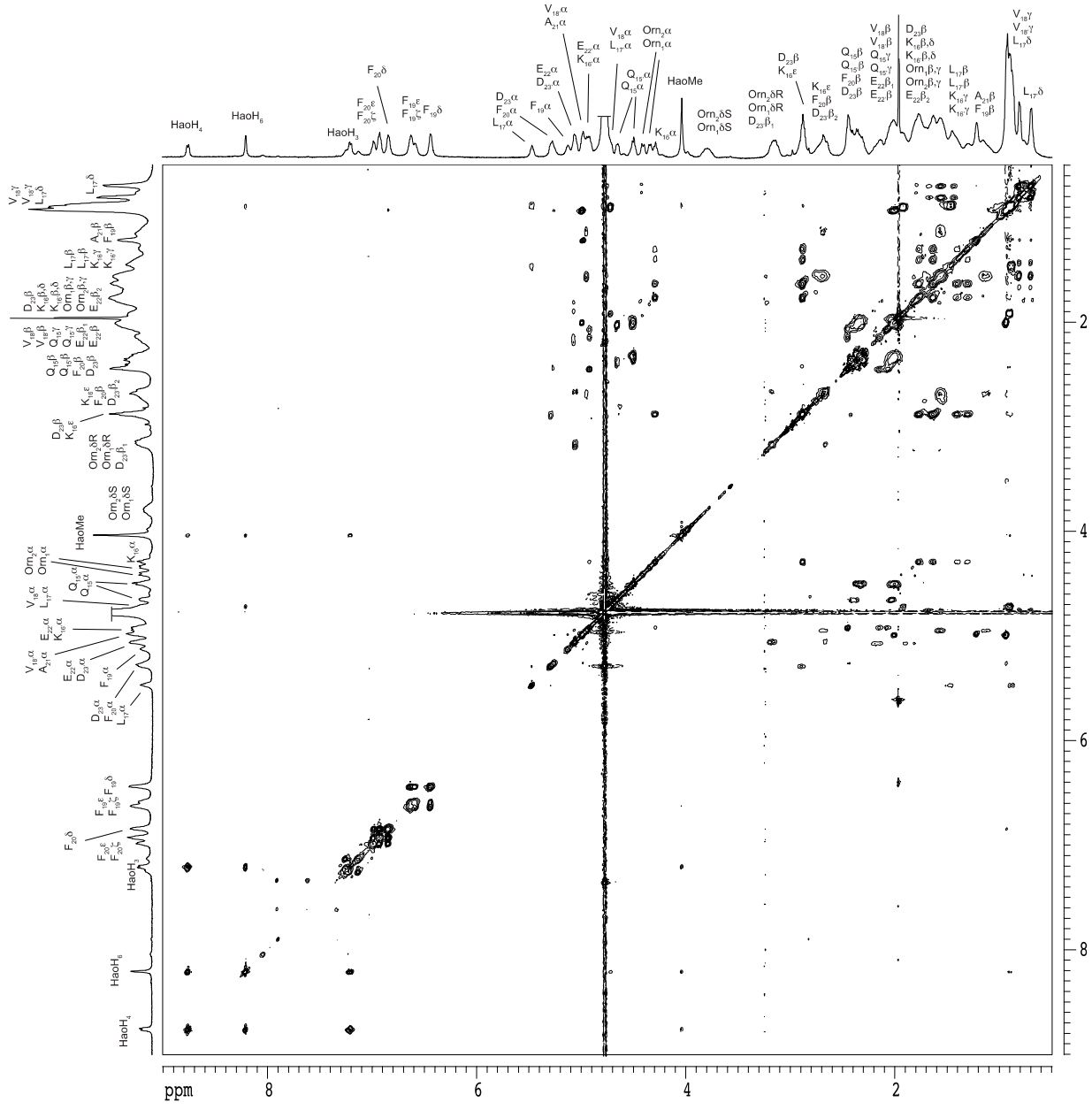
Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



2D TOCSY spectrum of macrocyclic β-sheet **4**
2 mM in D_2O, 500 MHz, 298 K
150-ms spin-locking mixing time
tetramer predominates

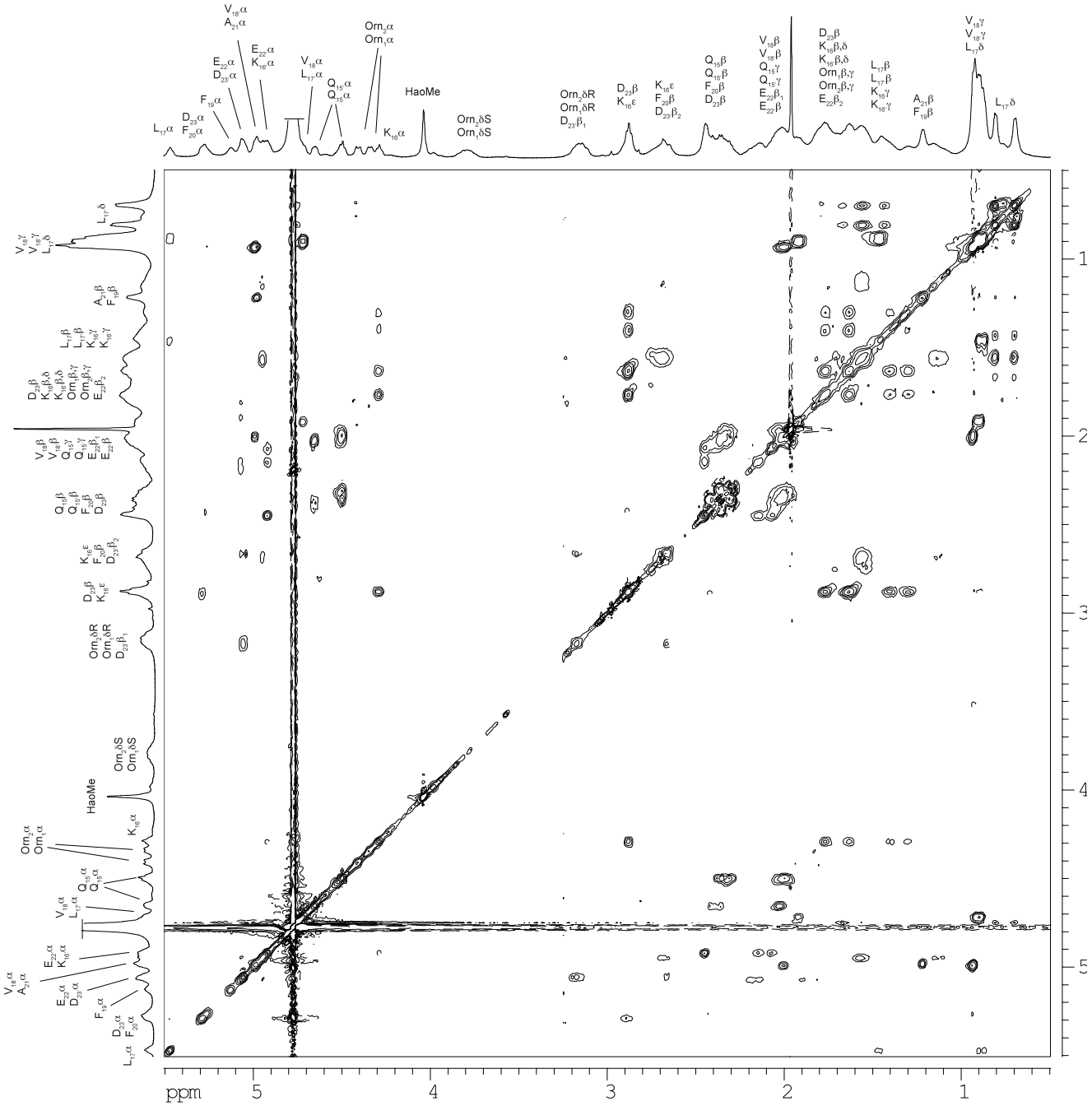
Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary





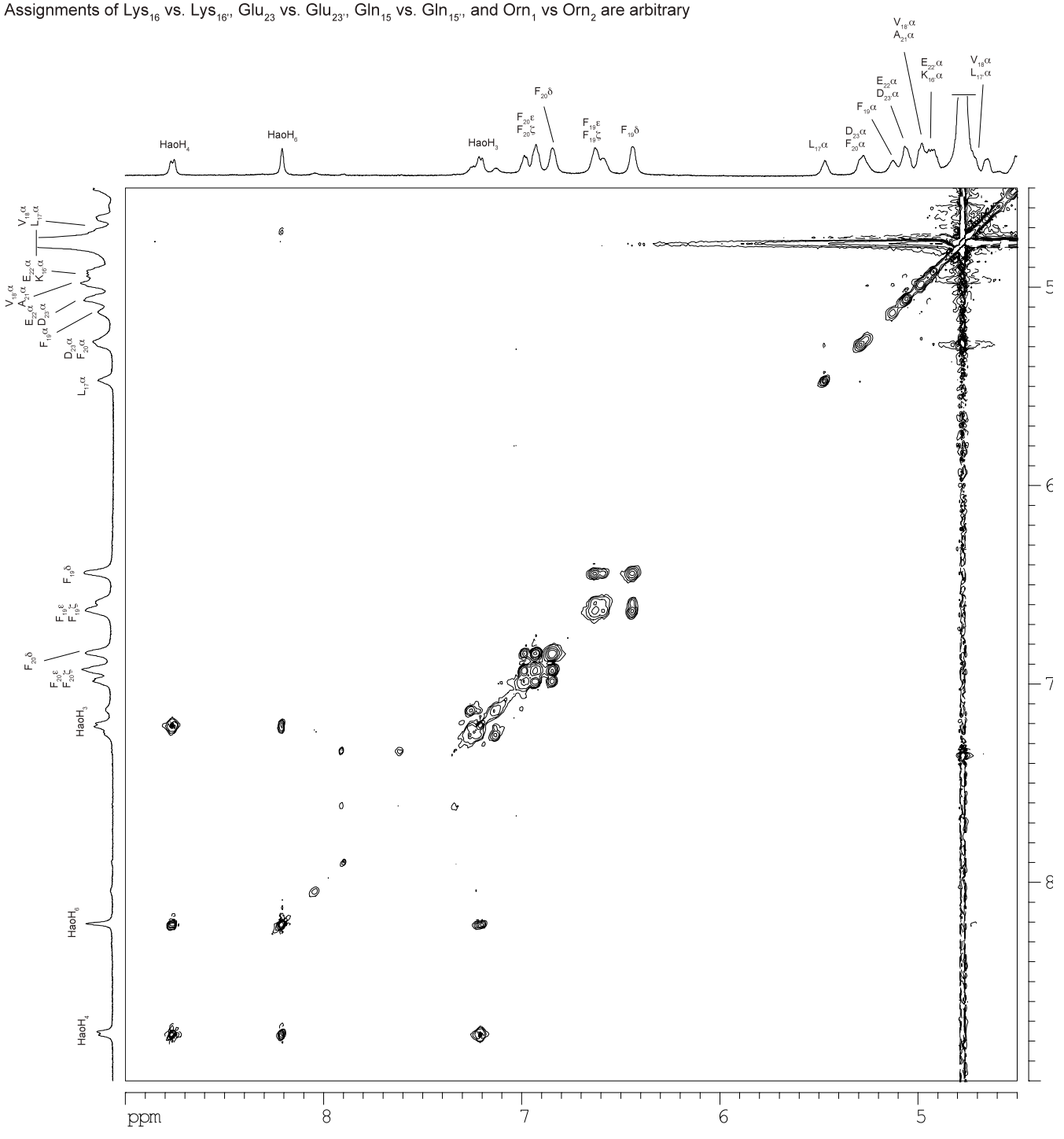
2D TOCSY spectrum of macrocyclic β-sheet **4**
2 mM in D_2O, 500 MHz, 298 K, 150-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



2D TOCSY spectrum of macrocyclic β-sheet **4**
2 mM in D_2O, 500 MHz, 298 K, 150-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



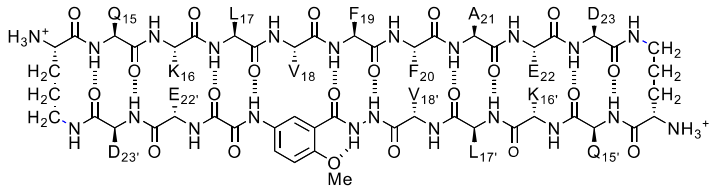
2D NOESY spectrum of macrocyclic β-sheet **4**

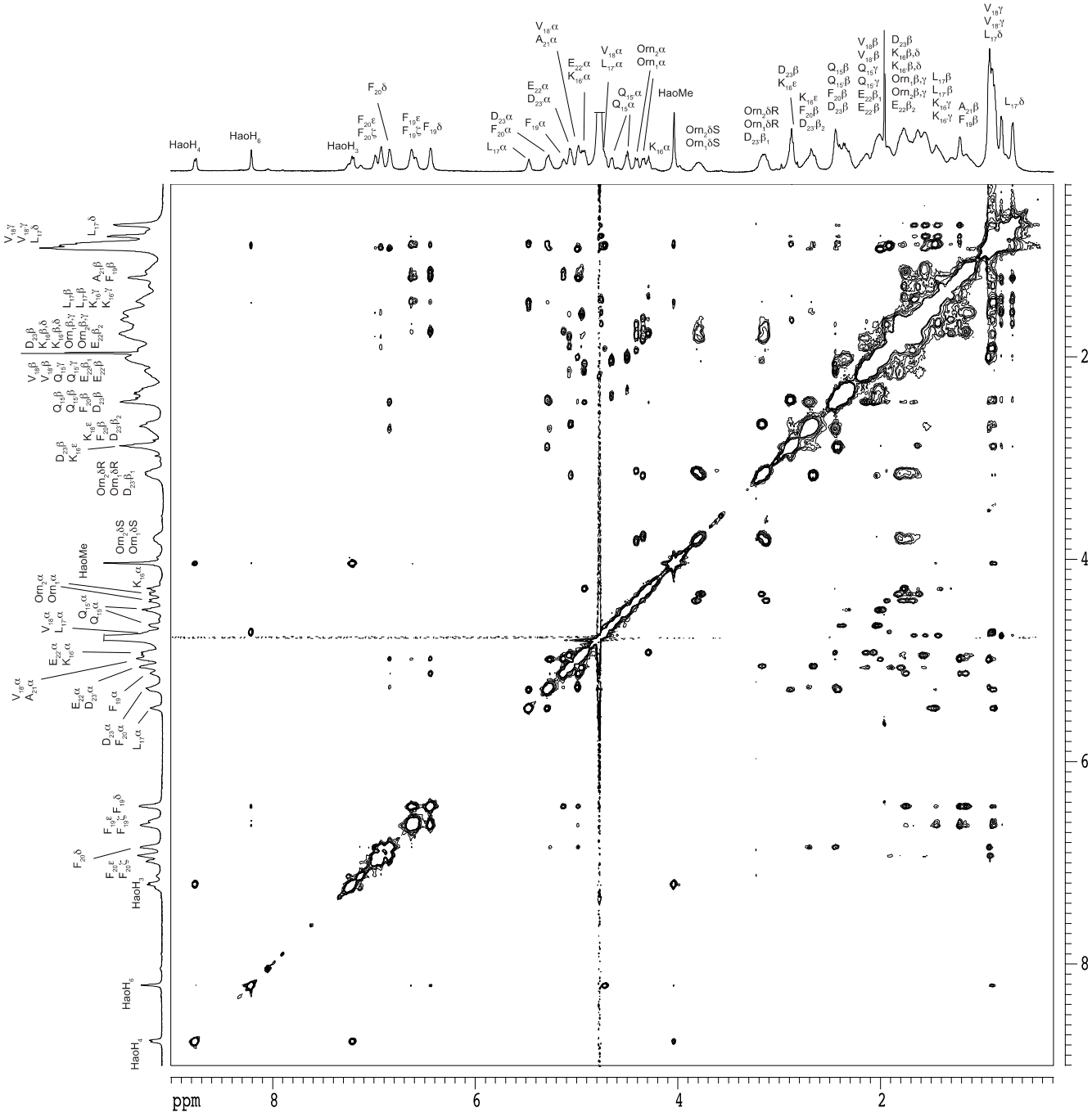
2 mM in D_2O, 500 MHz, 298 K

200-ms spin-locking mixing time

tetramer predominates

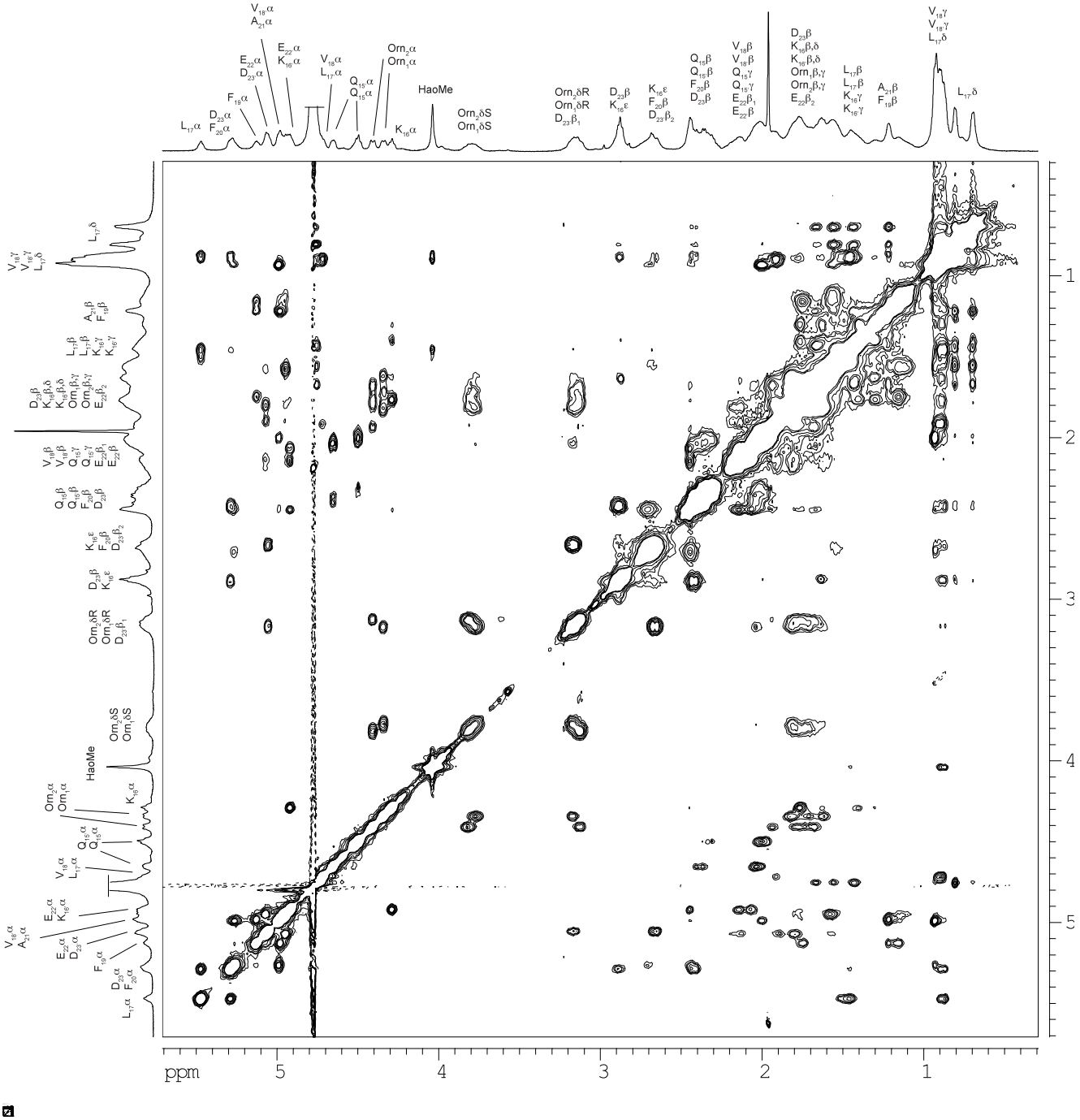
Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary





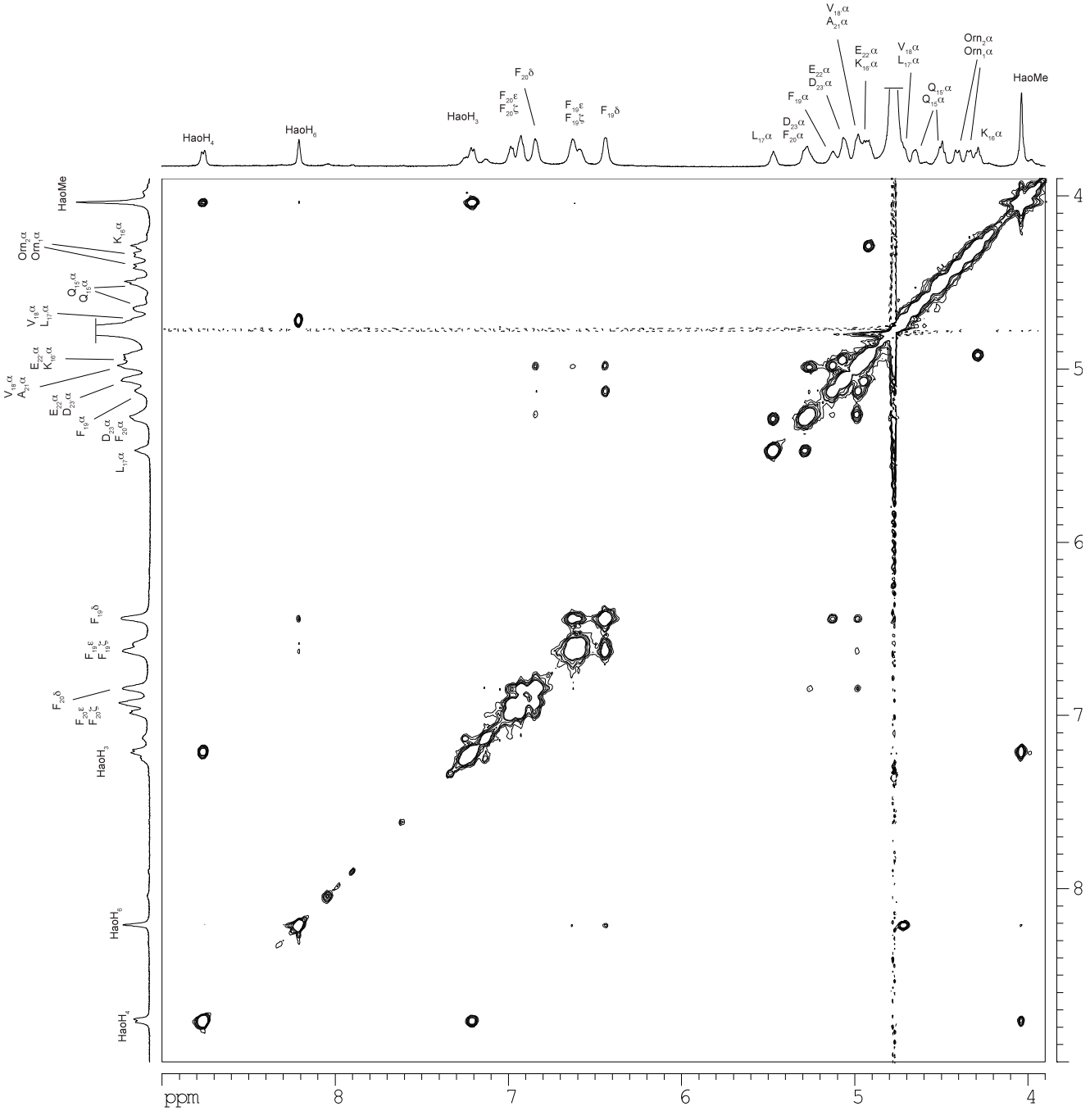
2D NOESY spectrum of macrocyclic β-sheet **4**
2 mM in D_2O, 500 MHz, 298 K, 200-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



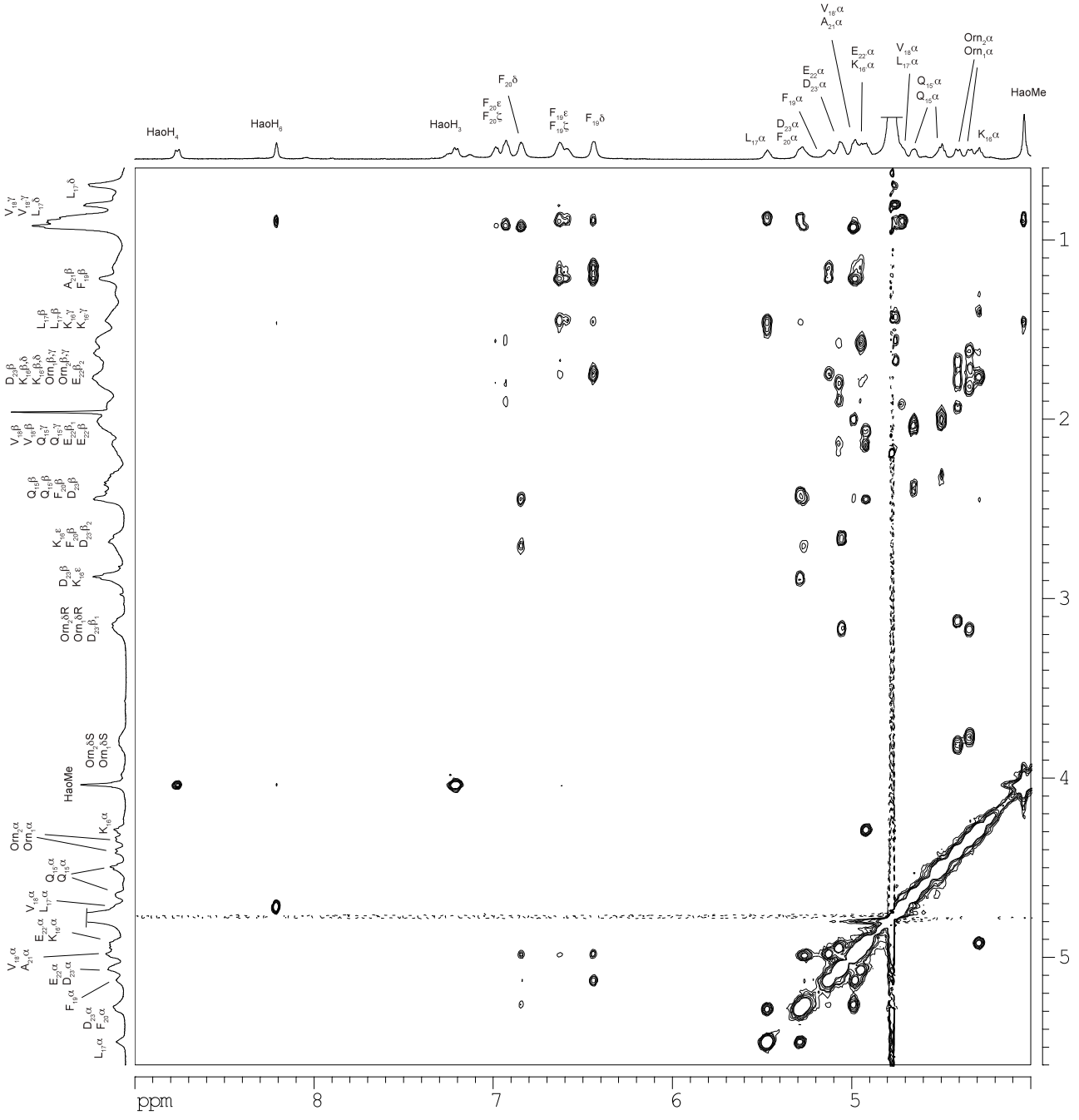
2D NOESY spectrum of macrocyclic β-sheet **4**
2 mM in D_2O, 500 MHz, 298 K, 200-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary

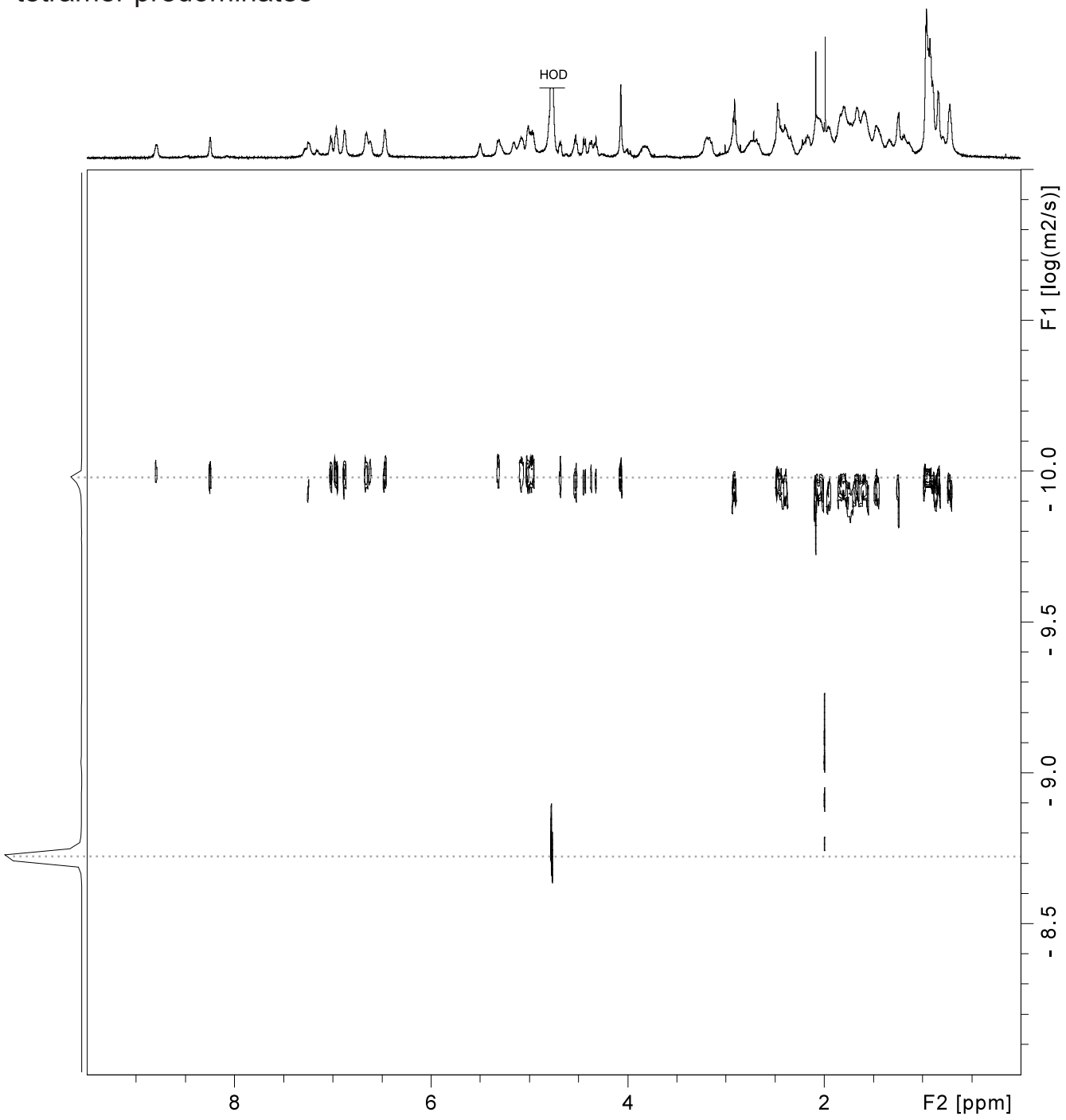


2D NOESY spectrum of macrocyclic β-sheet **4**
2 mM in D_2O, 500 MHz, 298 K, 200-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



2D DOSY spectrum of macrocyclic β-sheet **4**
2 mM in D_2O, 600 MHz, 298 K
tetramer predominates

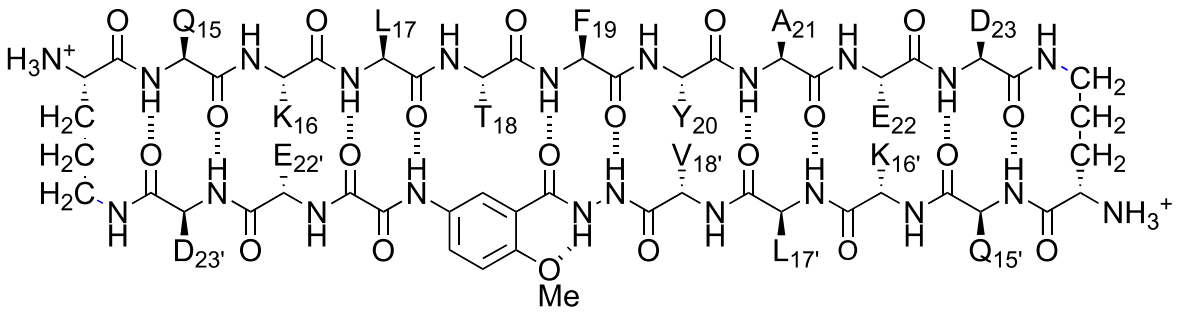


Calculation for peptide **4** at 2.0 mM

DC_{HOD} = 19.0 x 10^{-10} m^2/s [a]
log DC_{HOD} = -8.721

For peptide **4** tetramer, log DC (m^2/s) = -9.98(8), DC = $10^{-9.988}$ m^2/s = 10.3 x 10^{-11} m^2/s = 10.3 x 10^{-7} cm^2/s

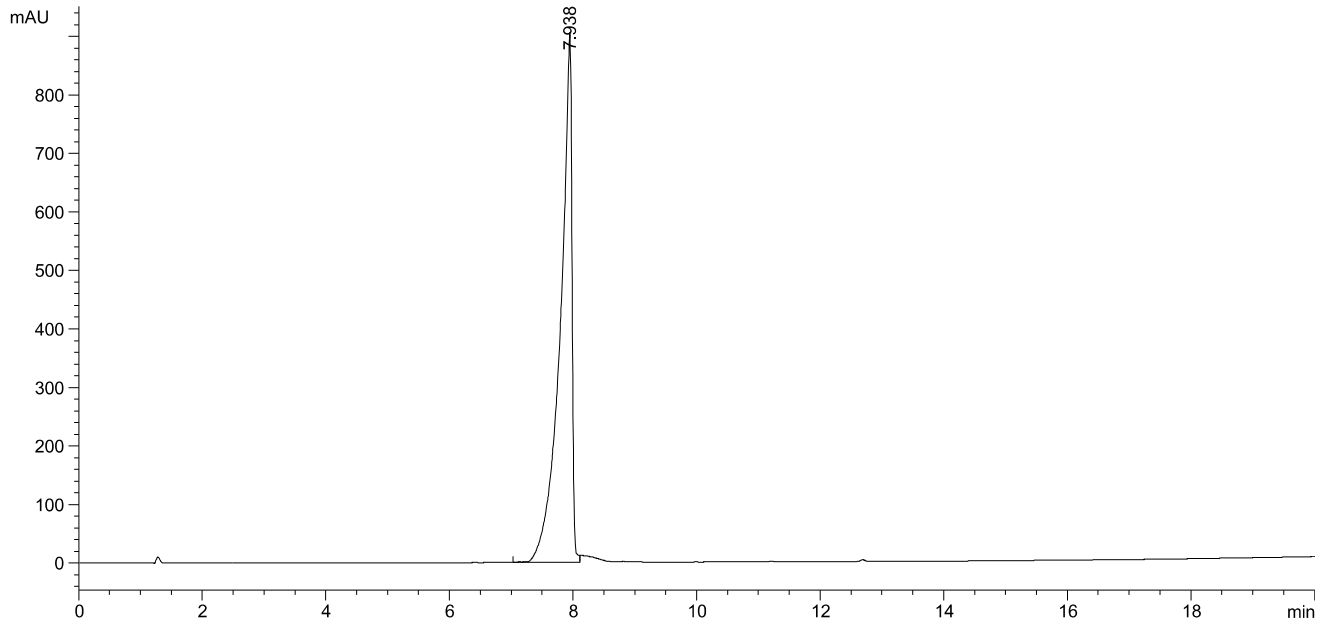
[a] Longsworth, L. G. J. Phys. Chem. 1960, 64, 1914–1917.



macrocyclic β-sheet peptide **5** (as the TFA salt)

molecular weight calculated for $C_{102}H_{154}N_{26}O_{33} \cdot 4CF_3CO_2H$ (TFA salt of **5**): 2728.56
molecular weight calculated for $C_{102}H_{154}N_{26}O_{33}$ (free base of **5**): 2272.47
exact mass calculated for $C_{102}H_{154}N_{26}O_{33}$ (free base of **5**): 2271.12

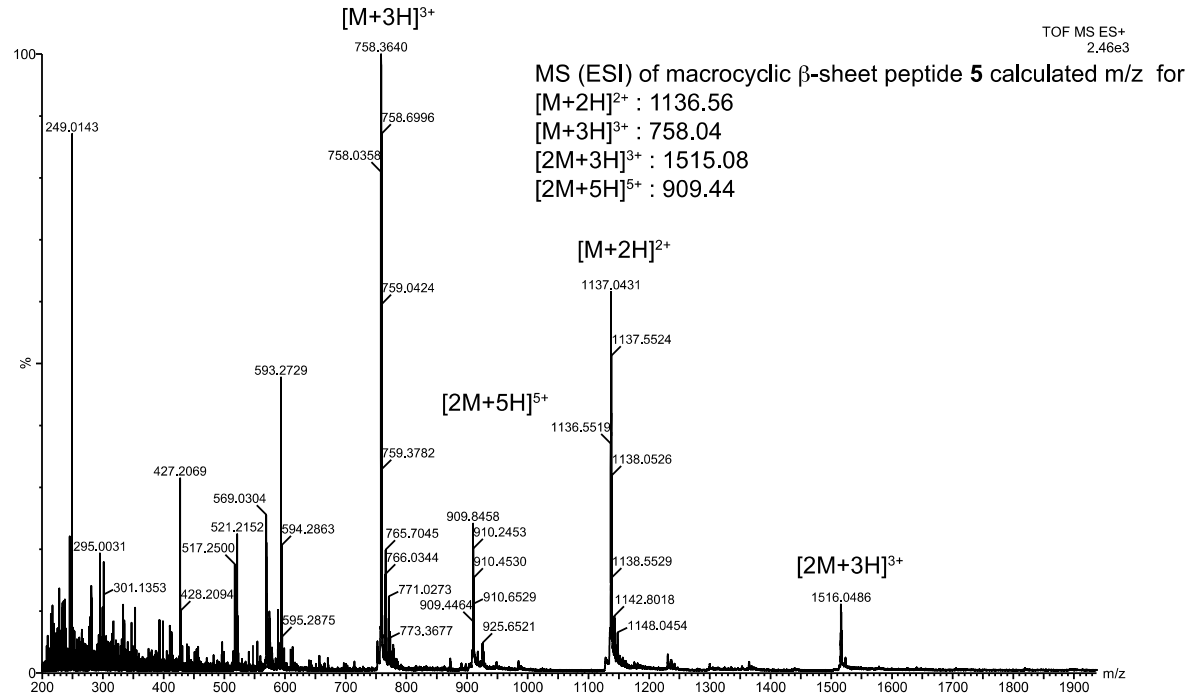
Analytical RP-HPLC of macrocyclic β-peptide **5**

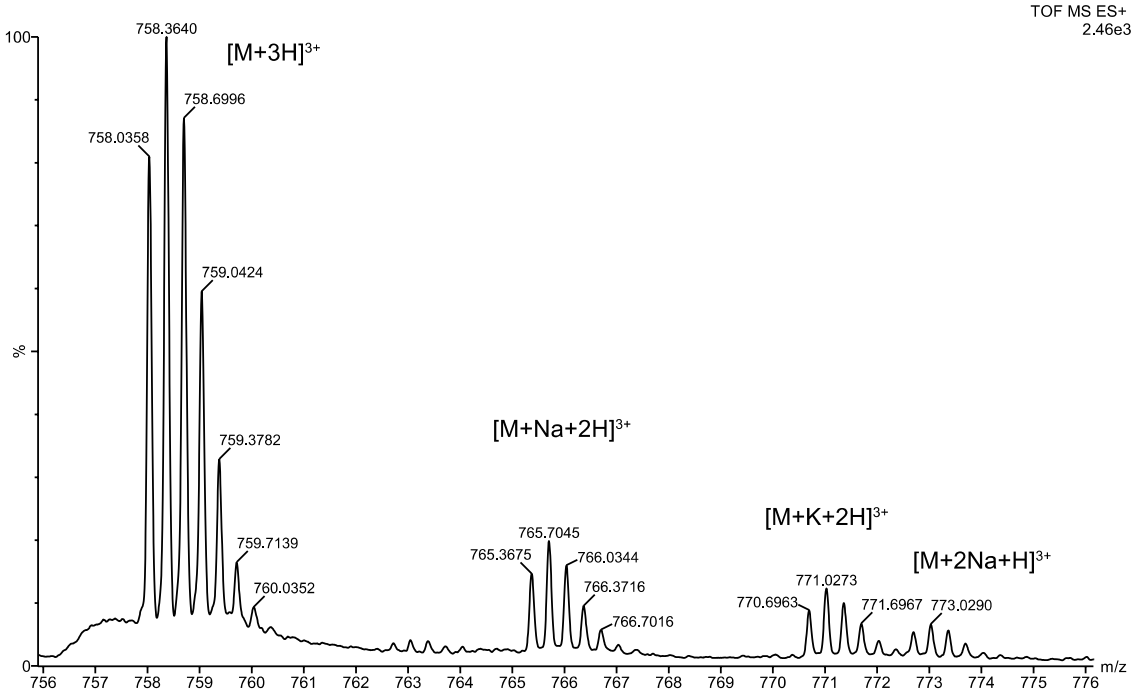


Signal 1: VWD1 A, Wavelength=254 nm

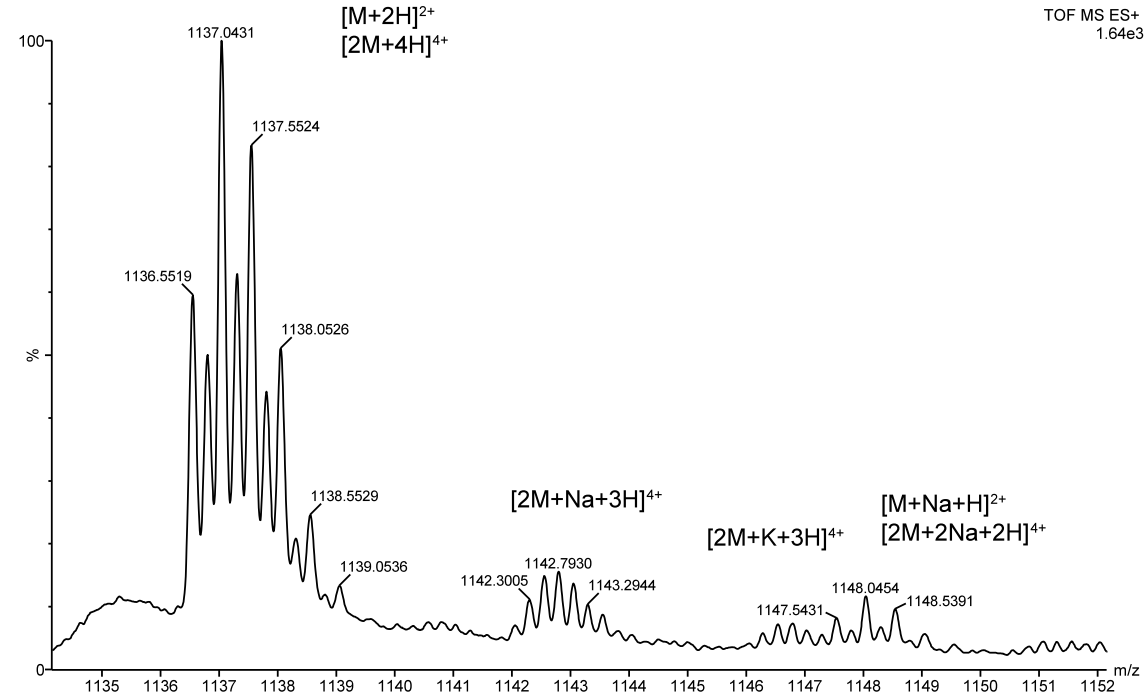
Peak #	RetTime [min]	Type	Width [min]	Area mAU *s	Height [mAU]	Area %
1	7.938	VV	0.1715	1.17436e4	906.91034	100.0000
Totals :				1.17436e4	906.91034	

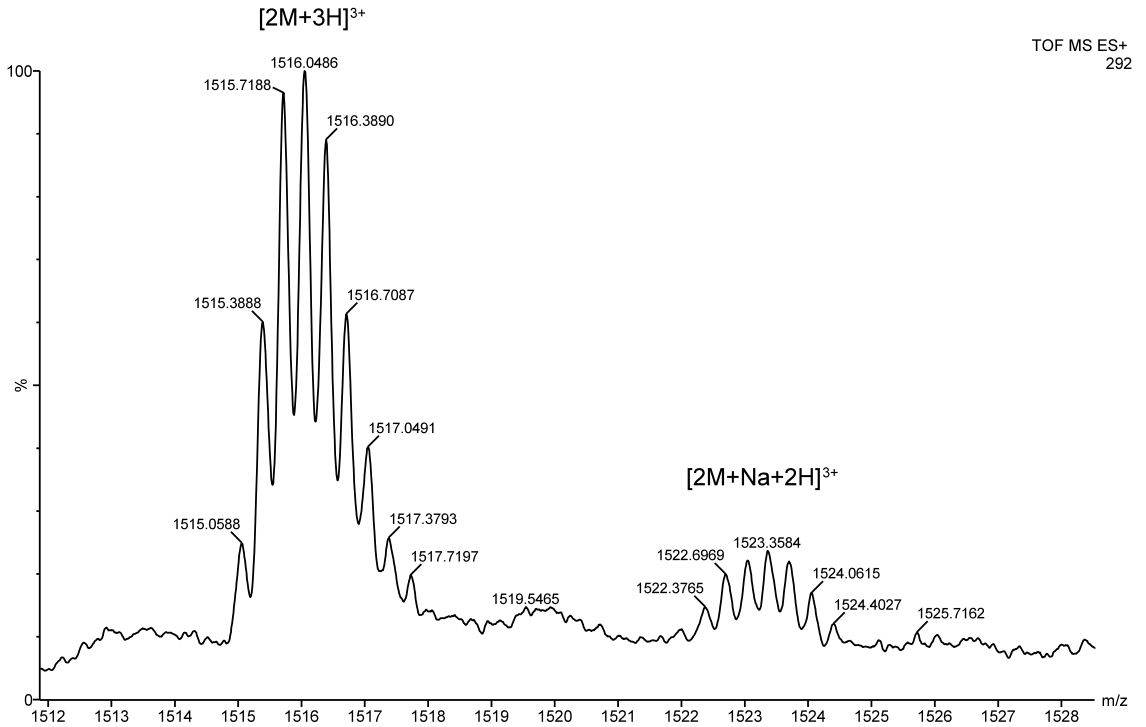
Macrocyclic β-sheet peptide **5**



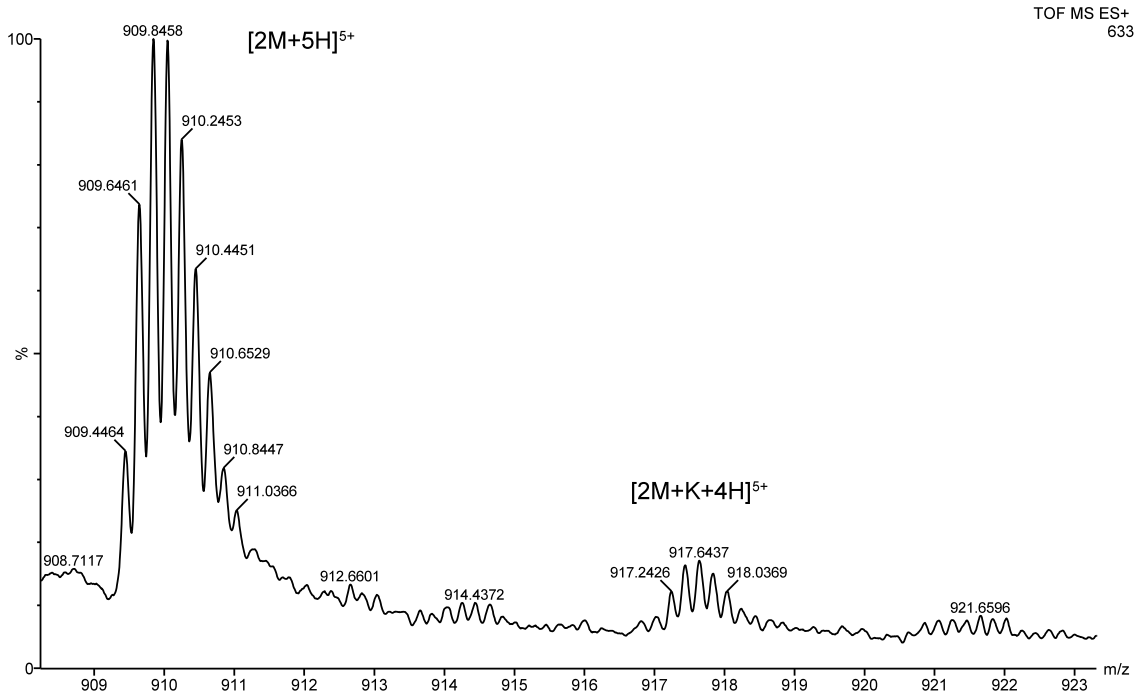


Macrocyclic β-sheet peptide **5**



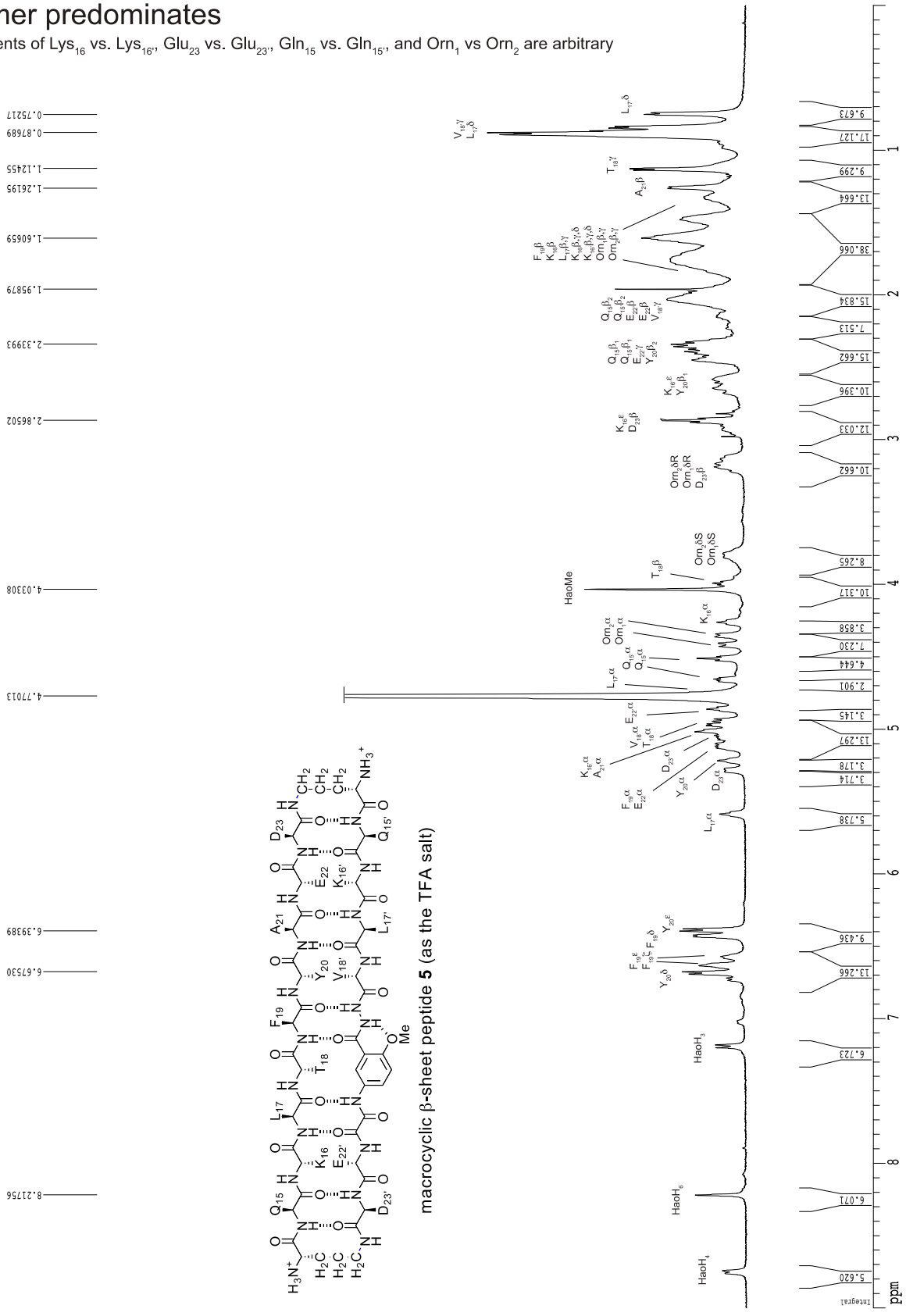


Macrocyclic β-sheet peptide **5**



1D ^{1}H NMR spectrum of macrocyclic β-sheet **5**
2 mM in D_2O, 500 MHz, 298 K
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



macrocyclic β-sheet peptide **5** (as the TFA salt)

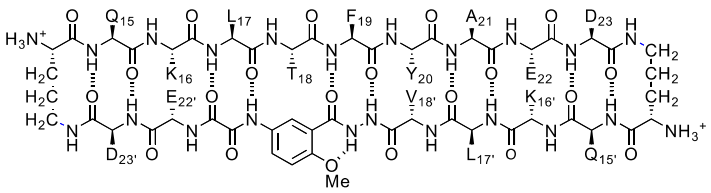
2D TOCSY spectrum of macrocyclic β-sheet **5**

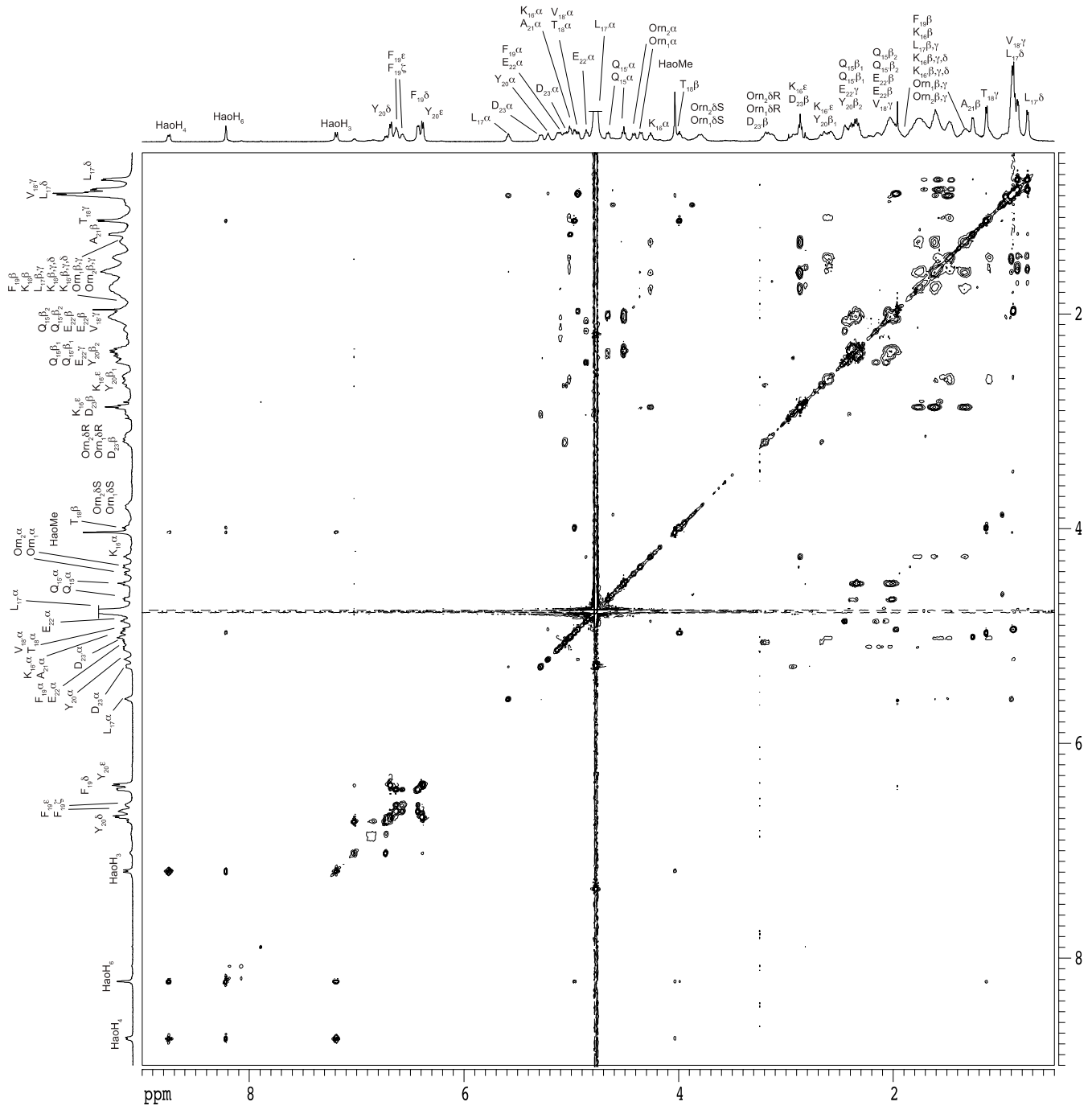
2 mM in D_2O, 500 MHz, 298 K

150-ms spin-locking mixing time

tetramer predominates

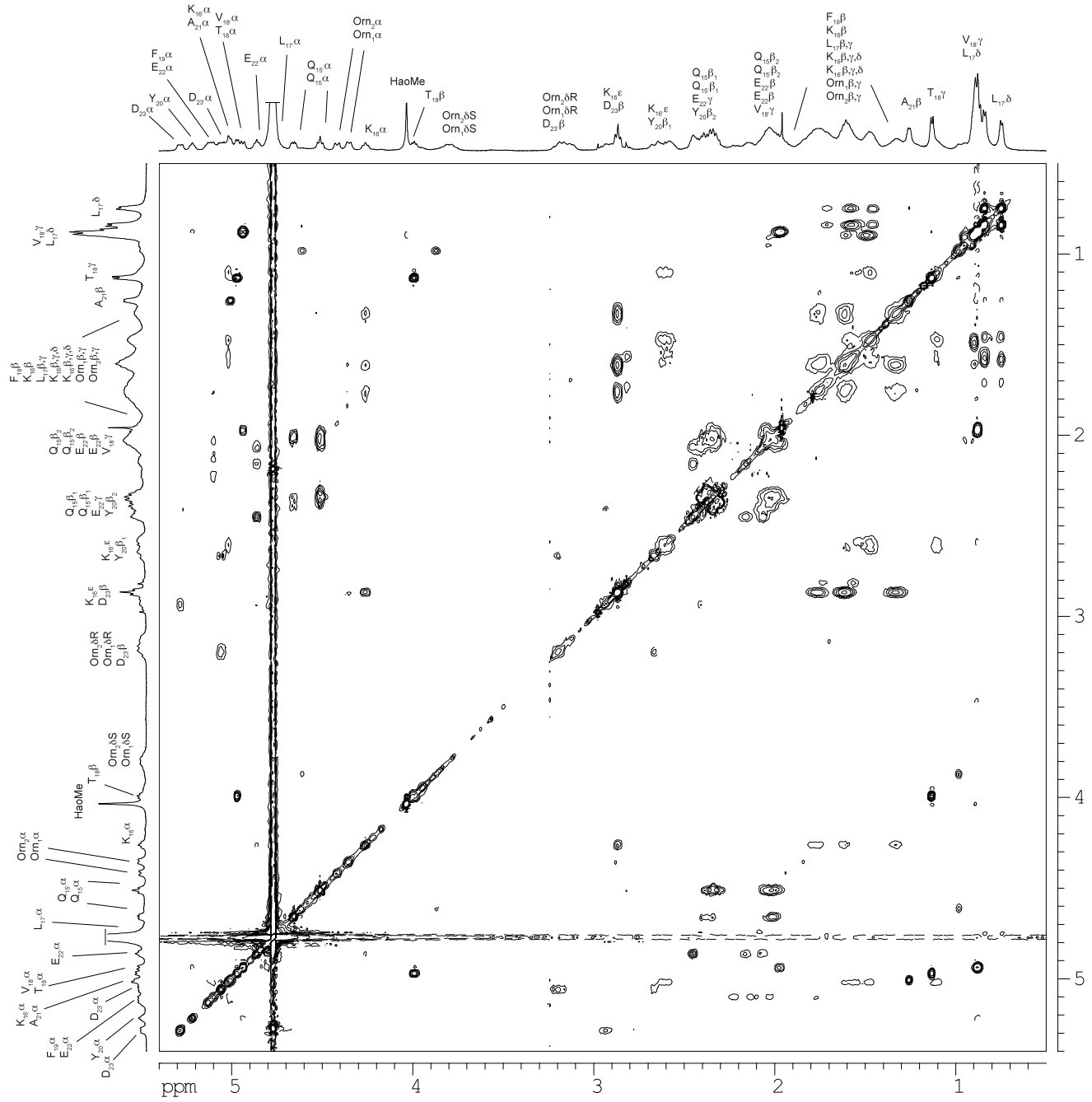
Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary





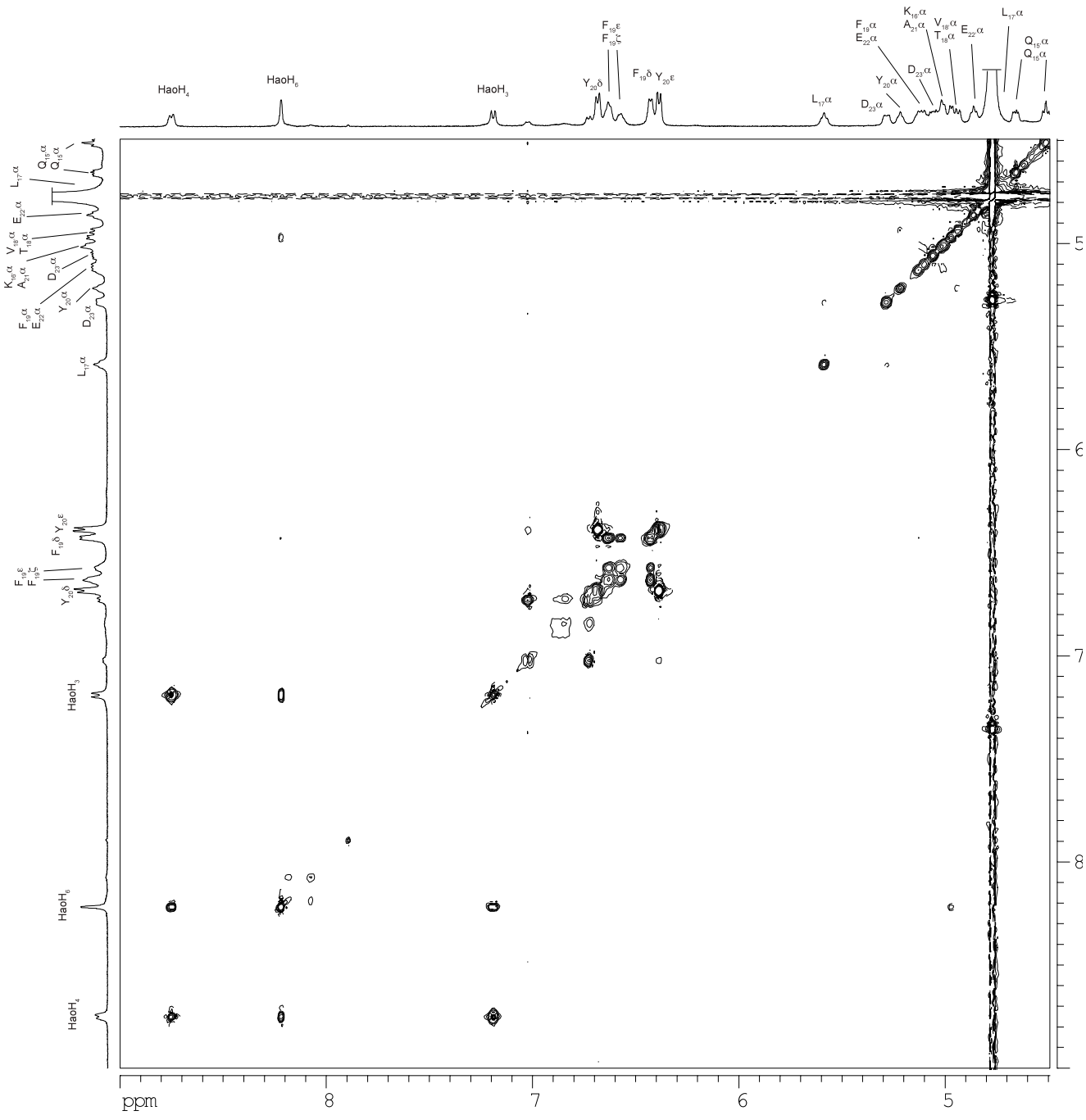
2D TOCSY spectrum of macrocyclic β-sheet **5**
2 mM in D_2O, 500 MHz, 298 K
150-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



2D TOCSY spectrum of macrocyclic β-sheet **5**
2 mM in D_2O, 500 MHz, 298 K
150-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



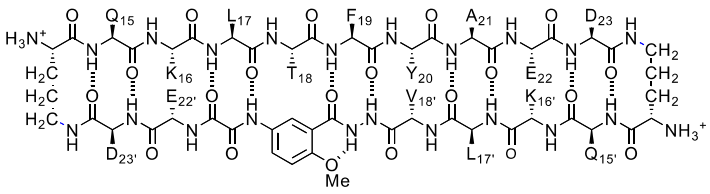
2D NOESY spectrum of macrocyclic β-sheet **5**

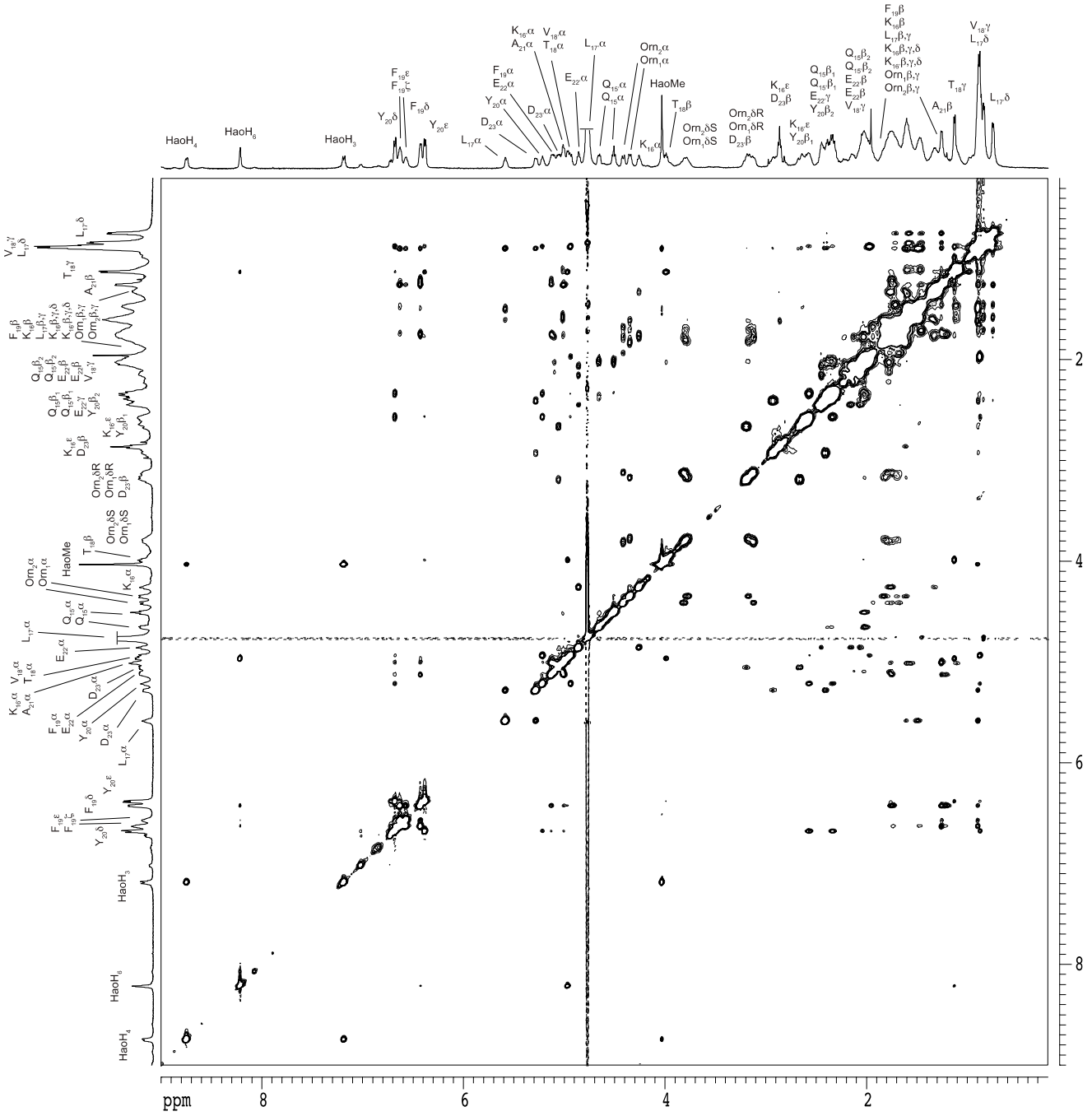
2 mM in D_2O, 500 MHz, 298 K

200-ms spin-locking mixing time

tetramer predominates

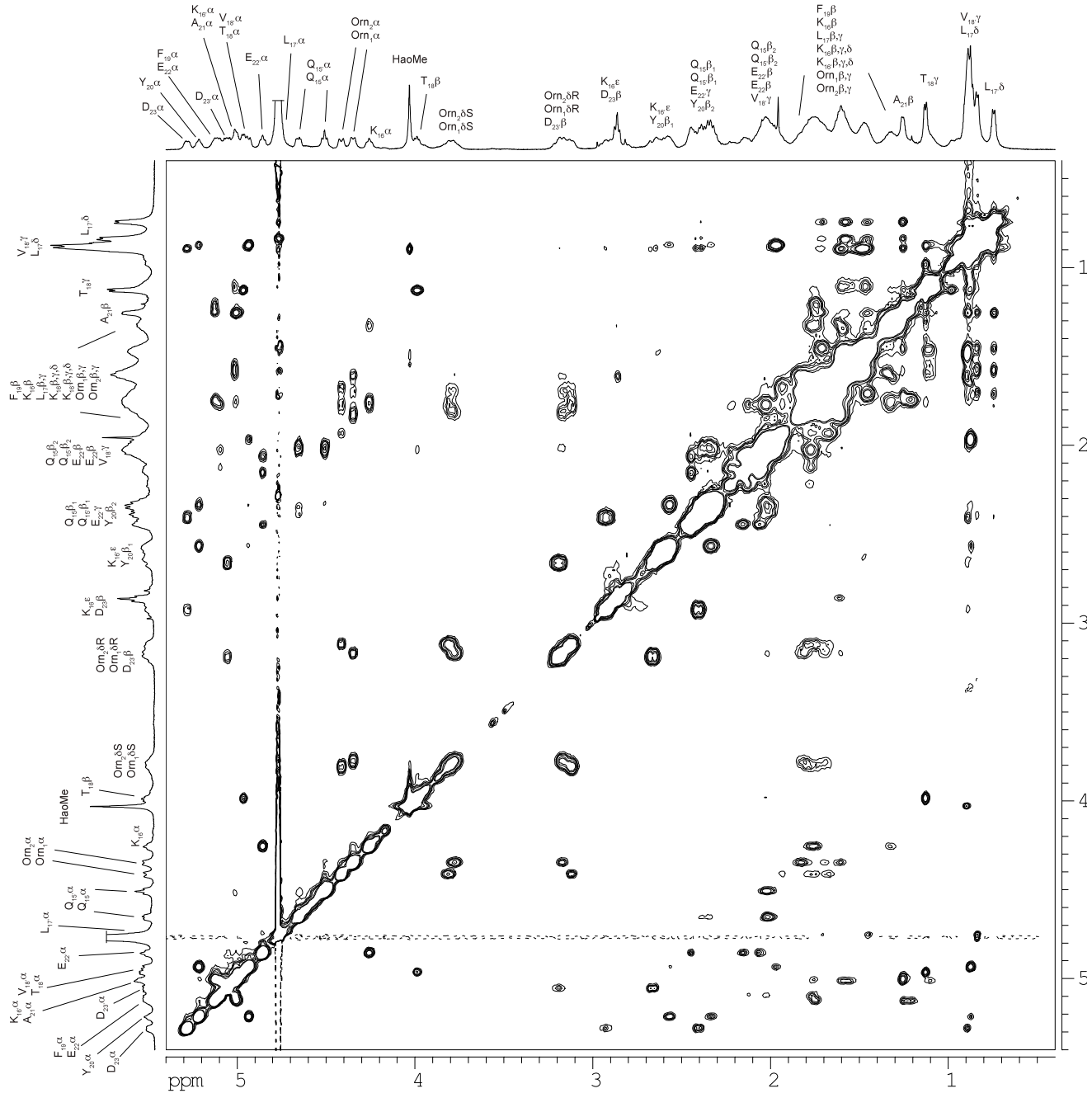
Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary





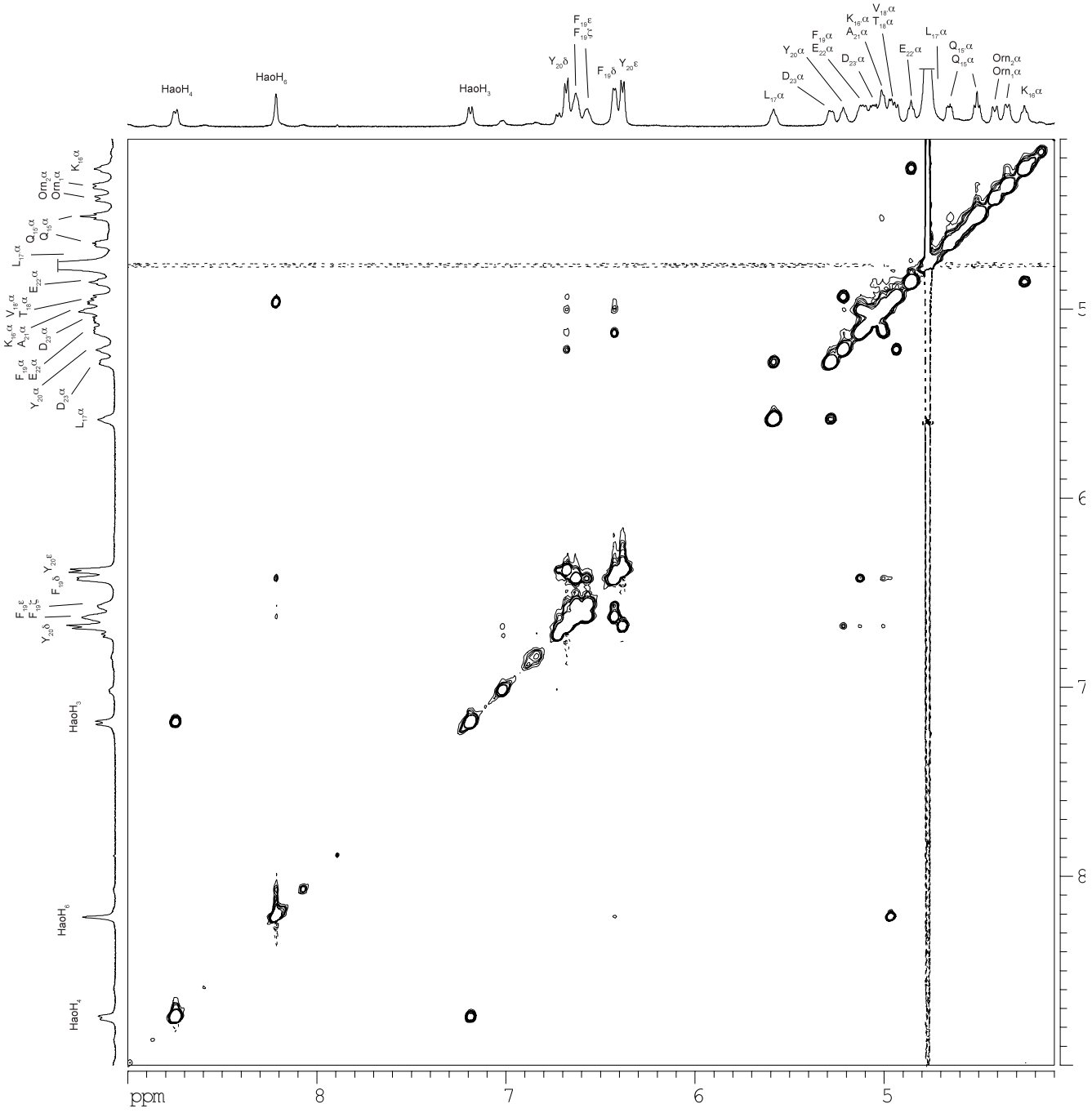
2D NOESY spectrum of macrocyclic β-sheet **5**
2 mM in D_2O, 500 MHz, 298 K
200-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



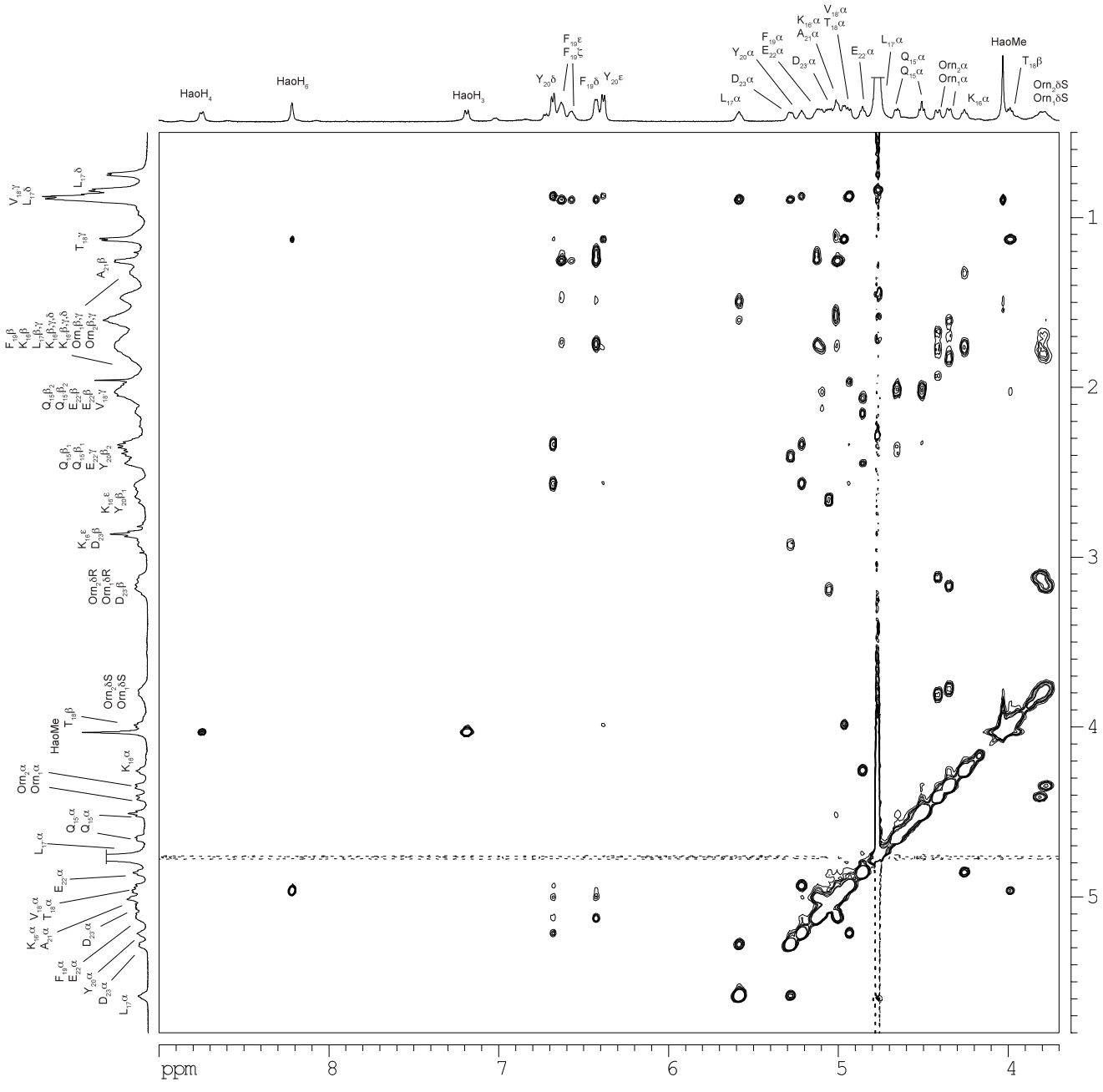
2D NOESY spectrum of macrocyclic β-sheet **5**
2 mM in D_2O, 500 MHz, 298 K
200-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary

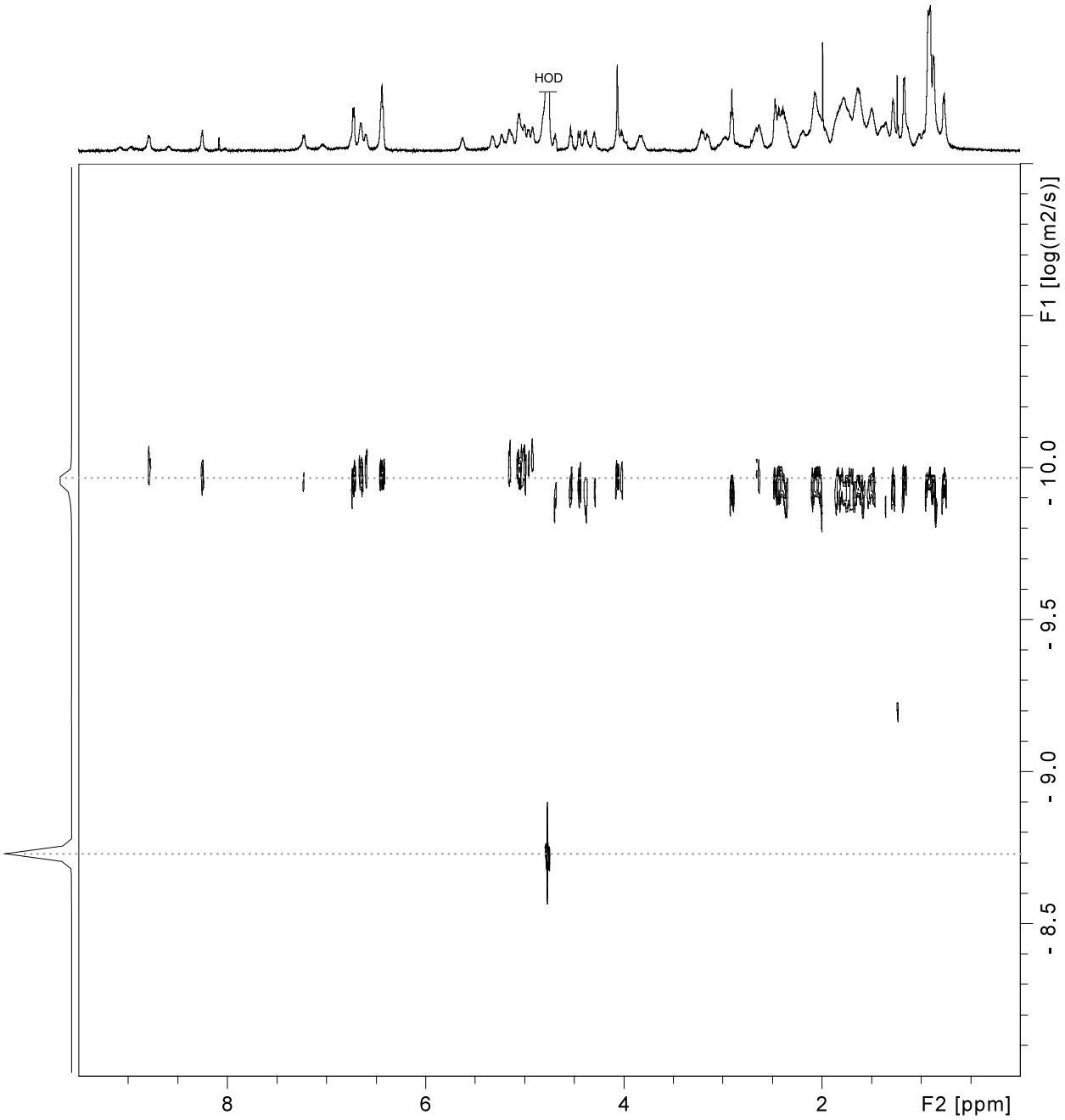


2D NOESY spectrum of macrocyclic β-sheet **5**
2 mM in D_2O, 500 MHz, 298 K
200-ms spin-locking mixing time
tetramer predominates

Assignments of Lys_{16} vs. $Lys_{16'}$, Glu_{23} vs. $Glu_{23'}$, Gln_{15} vs. $Gln_{15'}$, and Orn_1 vs Orn_2 are arbitrary



2D DOSY spectrum of macrocyclic β-sheet **5**
2 mM in D_2O, 600 MHz, 298 K
tetramer predominates

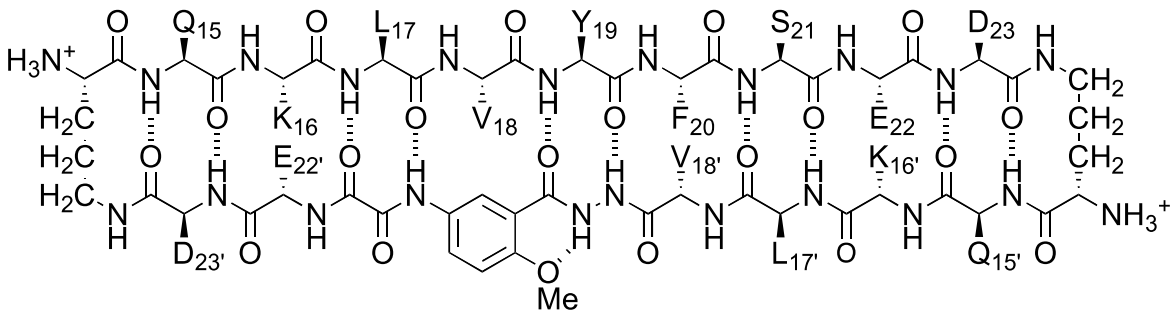


Calculation for peptide **5** at 2.0 mM

DC_{HOD} = 19.0 x 10^{-10} m^2/s [a]
log DC_{HOD} = -8.721

For peptide **5** tetramer, log DC (m^2/s) = -9.97(0), DC = $10^{-9.970}$ m^2/s = 10.7 x 10^{-11} m^2/s = 10.7 x 10^{-7} cm^2/s

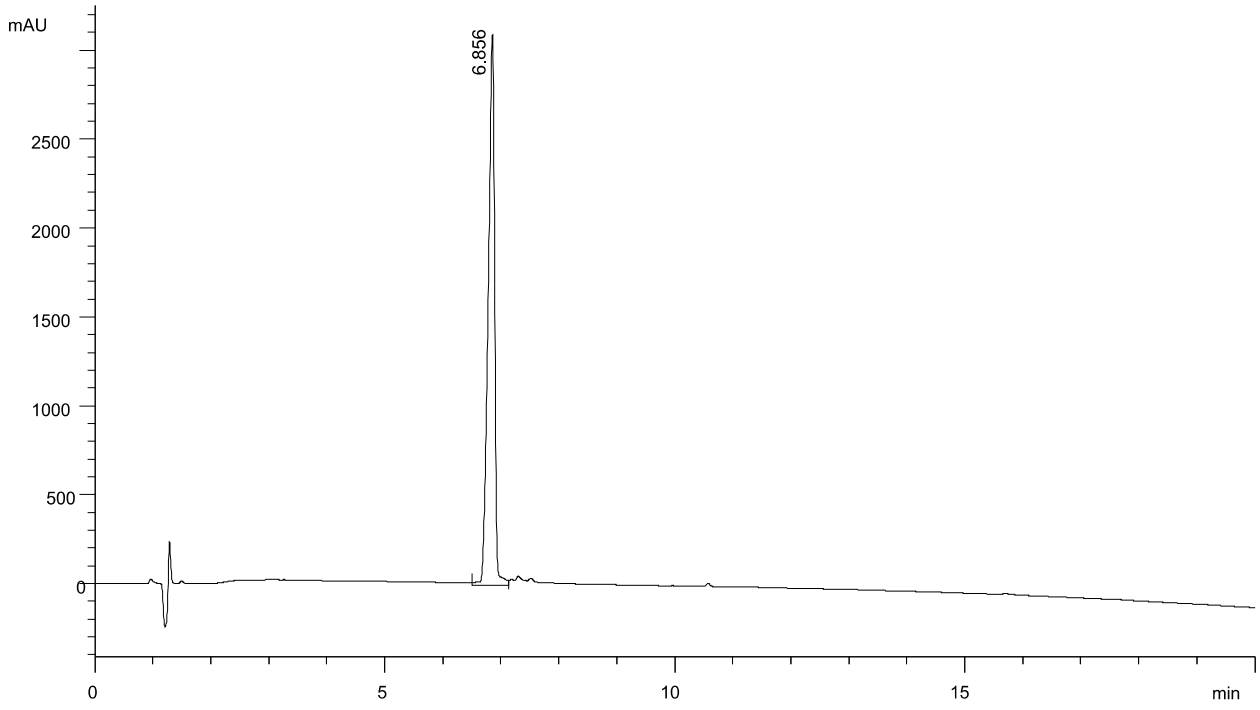
[a] Longsworth, L. G. J. Phys. Chem. 1960, 64, 1914–1917.



macrocyclic β-sheet peptide **6** (as the TFA salt)

molecular weight calculated for $C_{103}H_{156}N_{26}O_{33} \cdot 4CF_3CO_2H$ (TFA salt of **6**): 2742.59
molecular weight calculated for $C_{103}H_{156}N_{26}O_{33}$ (free base of **6**): 2286.50
exact mass calculated for $C_{103}H_{156}N_{26}O_{33}$ (free base of **6**): 2285.13

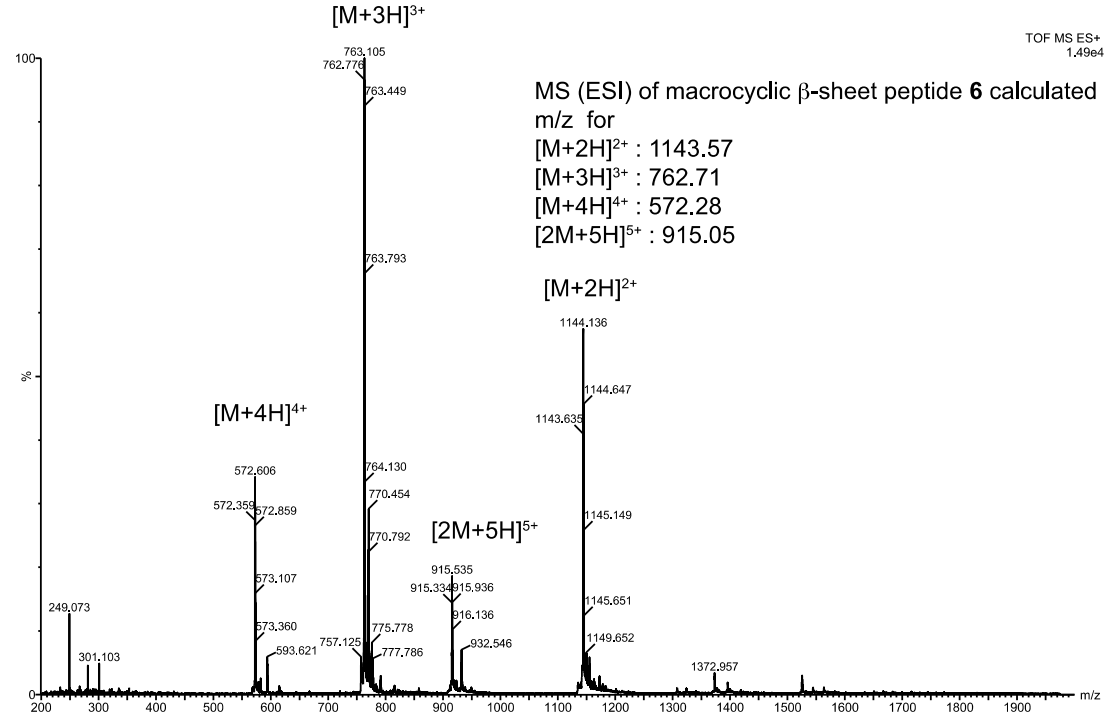
Analytical RP-HPLC of macrocyclic β-sheet **6**

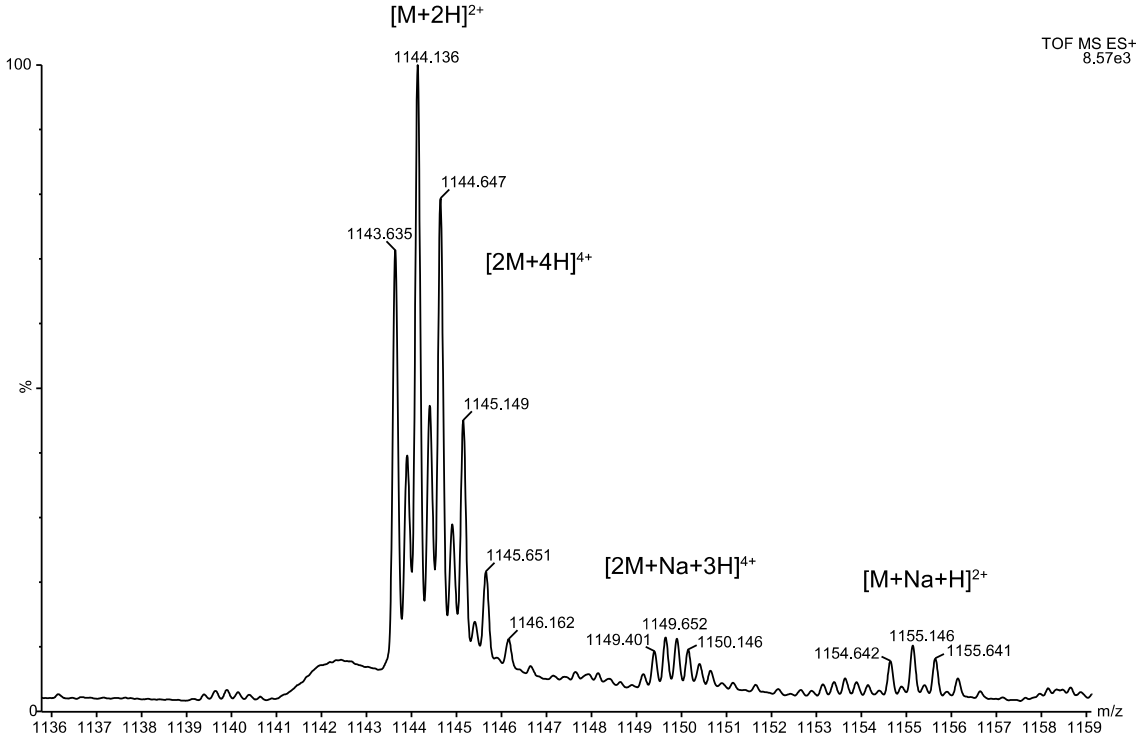


Signal 1:VWD1 A, Wavelength=214 nm

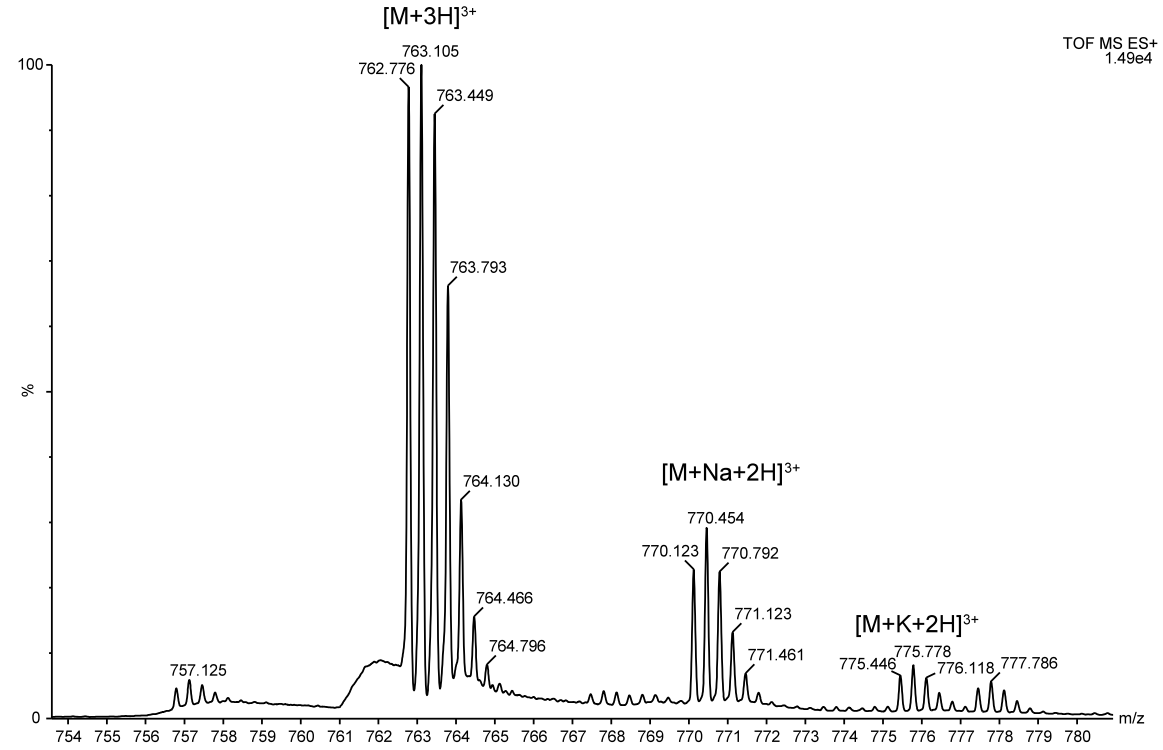
Peak #	RT [min]	Type	Width [min]	Area mAU*s	Height [mAU]	Area %
1	6.856	VV	0.108	23348.021	100.000	100.000

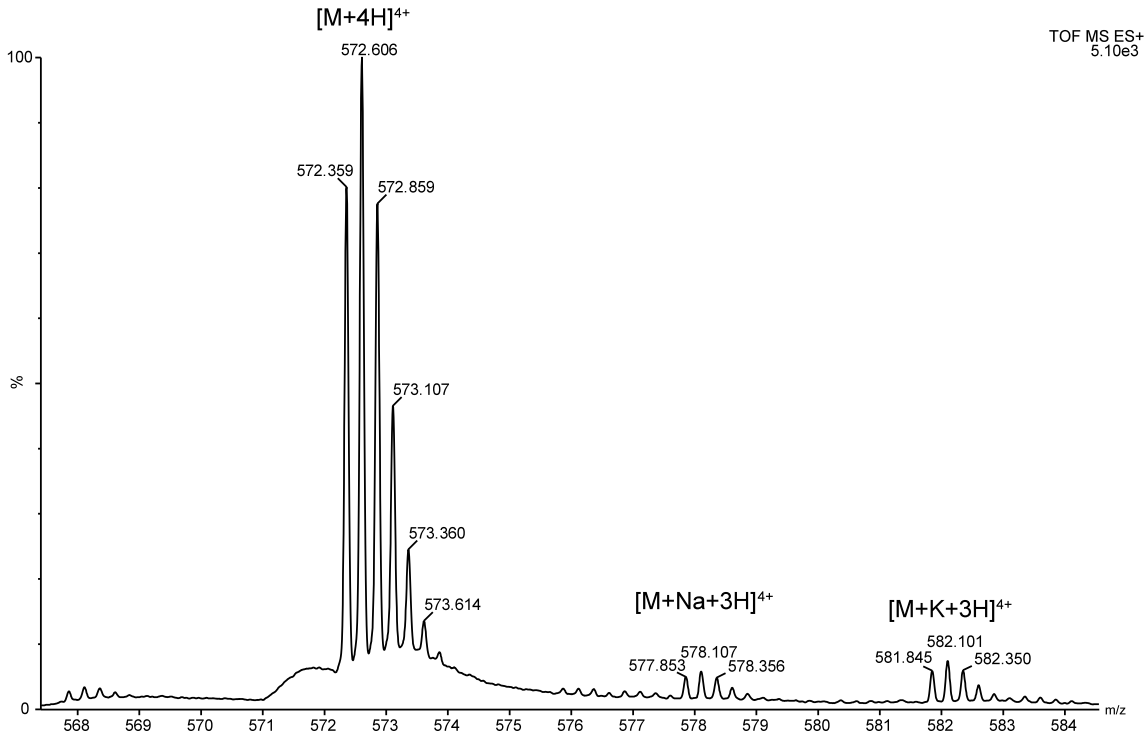
Macrocyclic β-sheet peptide **6**



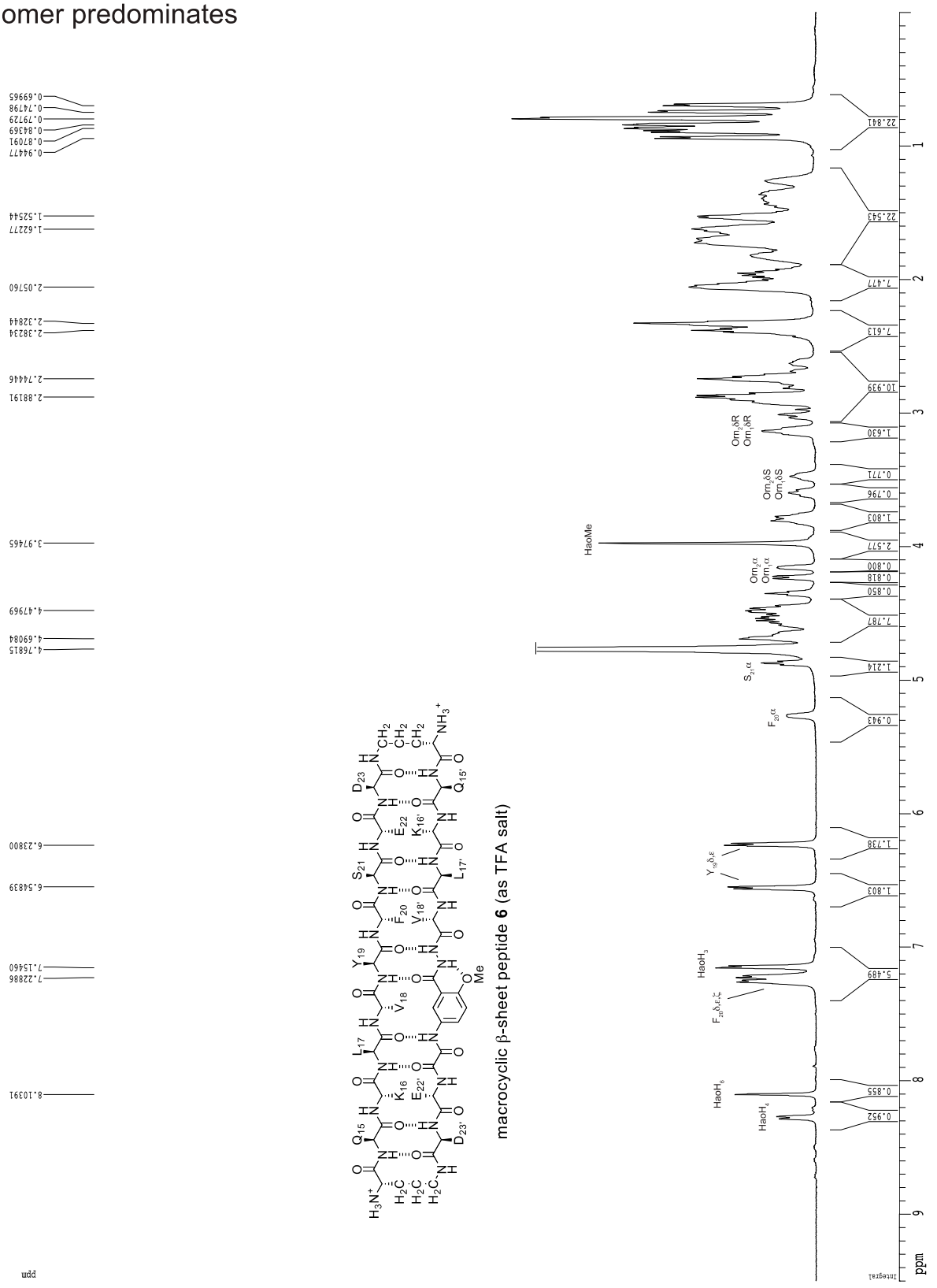


Macrocyclic β-sheet peptide **6**

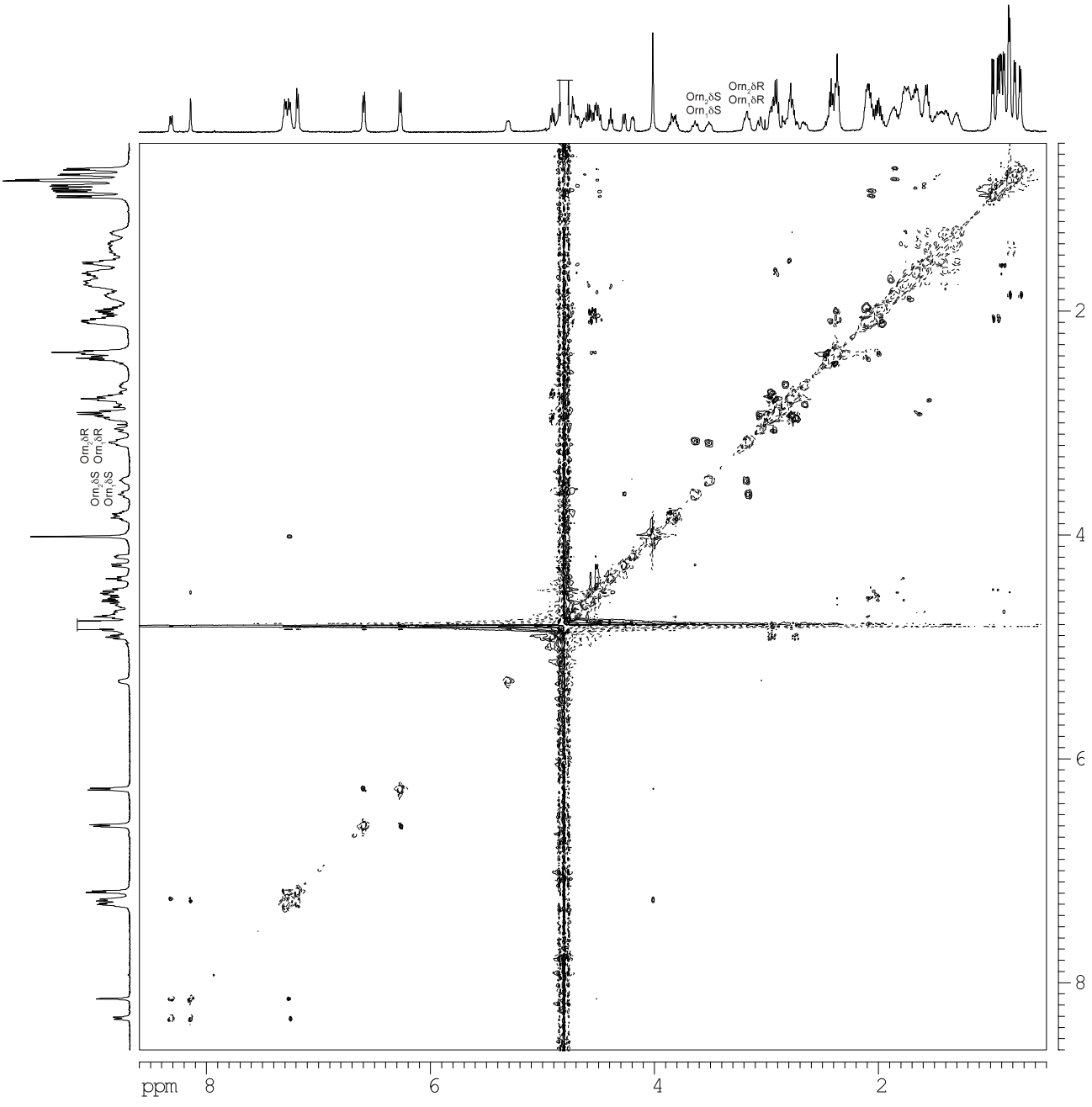




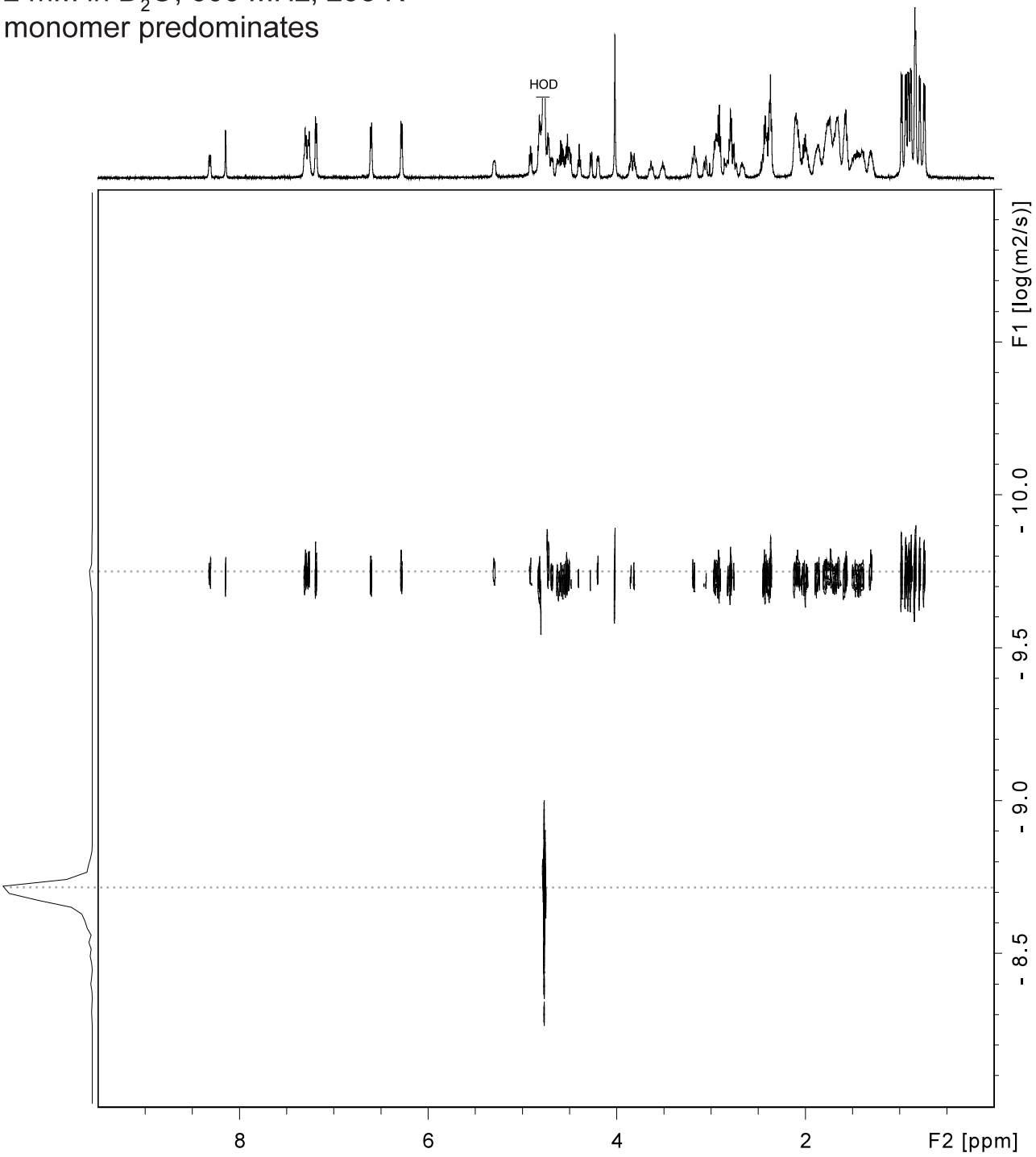
1D ^{1}H NMR spectrum of macrocyclic β-sheet **6**
2 mM in D_2O, 500 MHz, 298 K
monomer predominates



2D ROESY spectrum of macrocyclic β-sheet **6**
2 mM in D_2O, 500 MHz, 298 K
150-ms spin-lock mixing time
monomer predominates



2D DOSY spectrum of macrocyclic β-sheet **6**
2 mM in D_2O, 600 MHz, 298 K
monomer predominates



Calculation for peptide **6** at 2.0 mM

DC_{HOD} = 19.0 x 10^{-10} m^2/s [a]
log DC_{HOD} = -8.721

For peptide **6** tetramer, log DC (m^2/s) = -9.76(0), DC = $10^{-9.760}$ m^2/s = 17.4 x 10^{-11} m^2/s = 17.4 x 10^{-7} cm^2/s

[a] Longsworth, L. G. J. Phys. Chem. 1960, 64, 1914–1917.